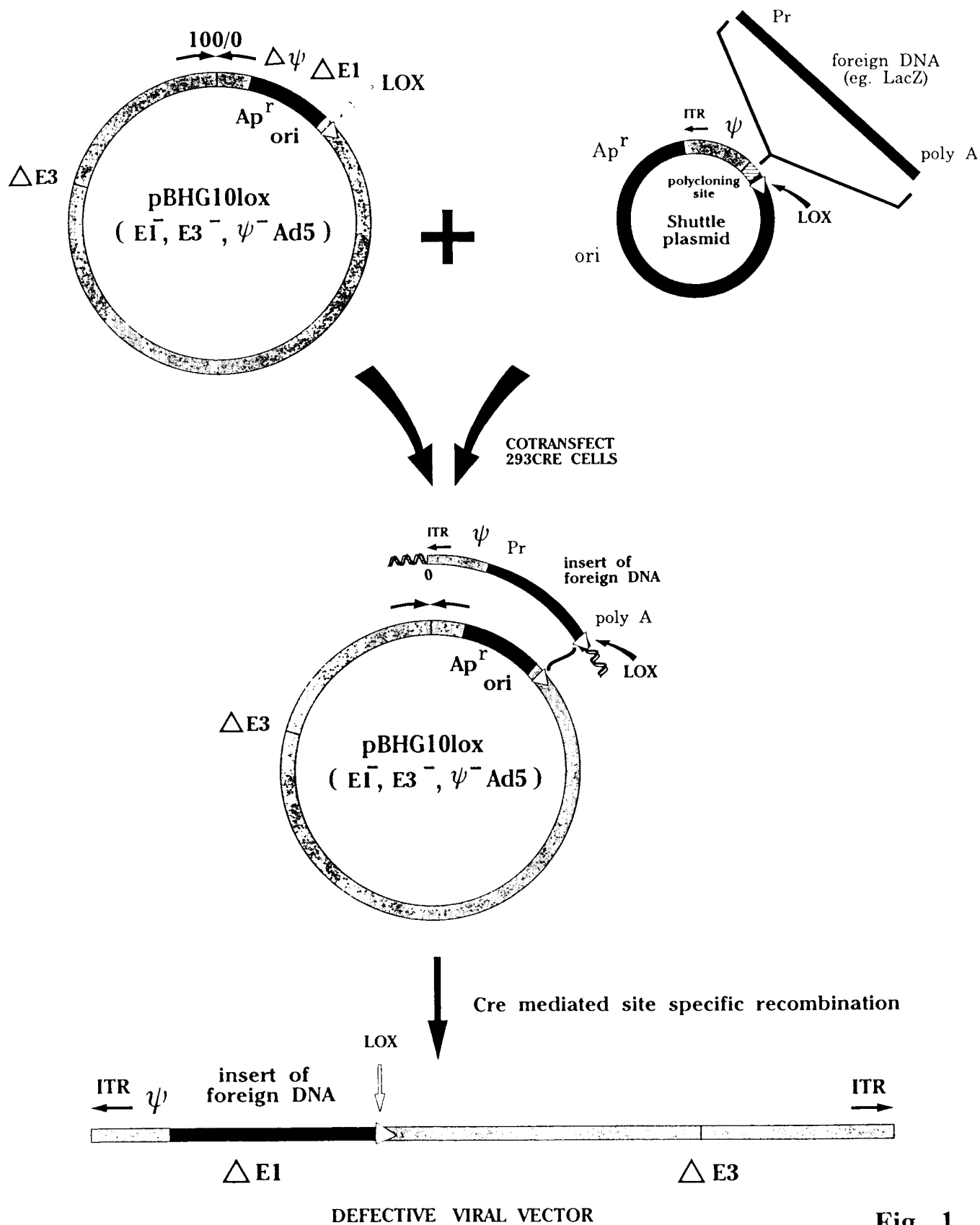
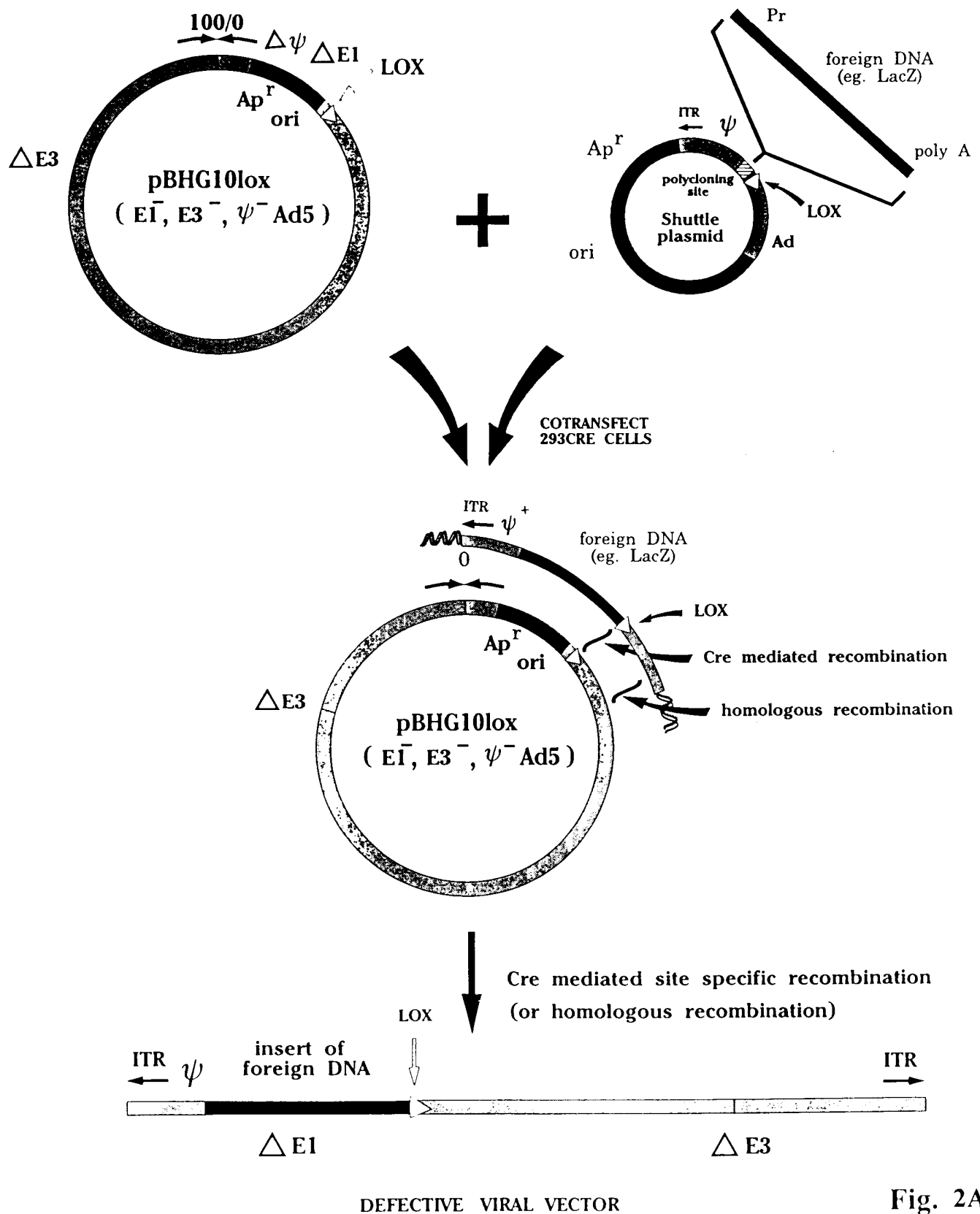


**Cotransfection of 293Cre cells with pBHG10lox and a "Lox" shuttle plasmid for generation of Ad expression vectors**



**Fig. 1**

**Cotransfection of 293Cre cells with pBHG10lox and a "lox" shuttle plasmid for generation of Ad expression vectors**



**Fig. 2A**

# CONSTRUCTION OF VARIOUS SHUTTLE PLASMIDS

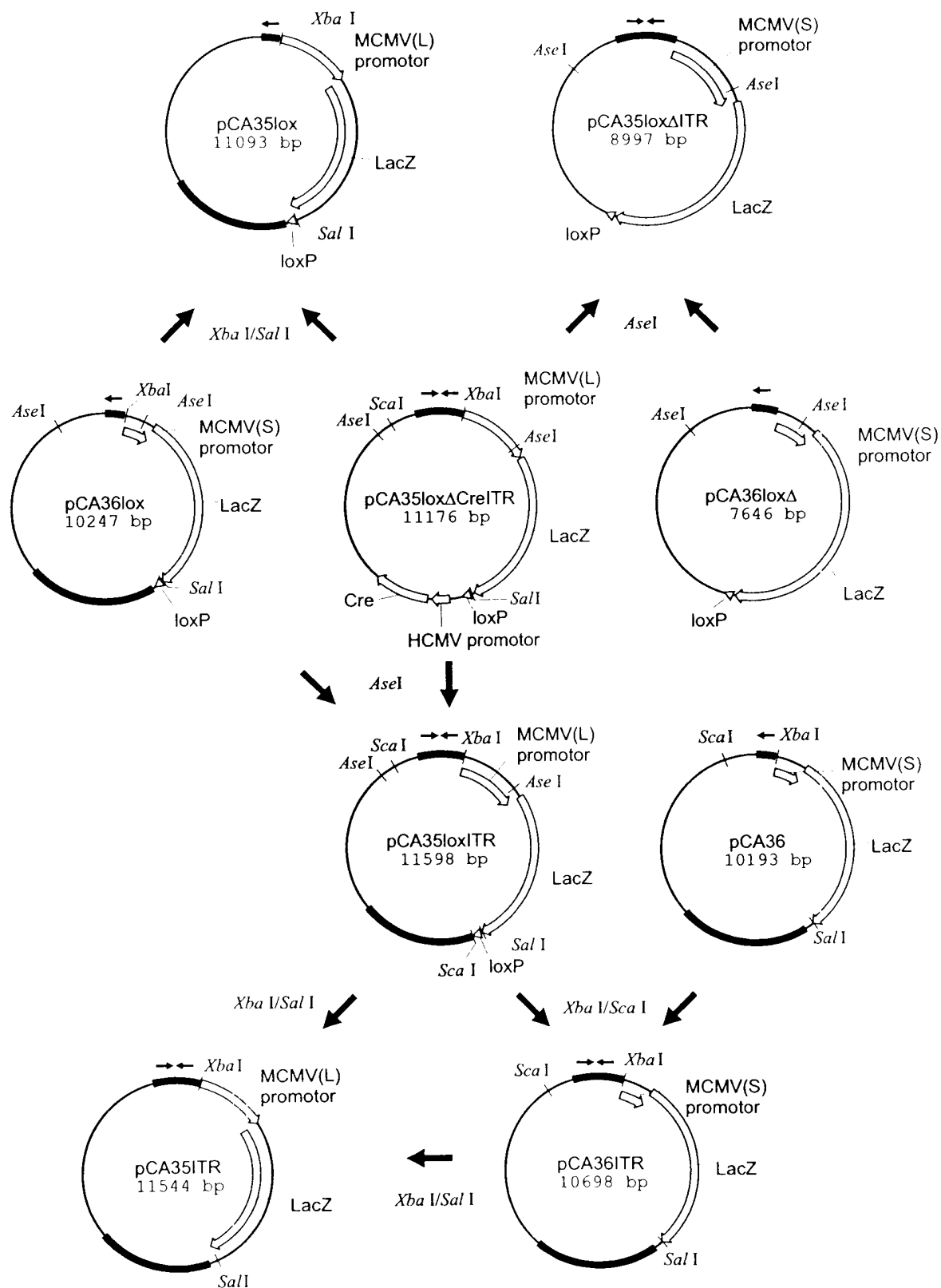


Figure 2B

# OLIGONUCLEOTIDES USED IN CLONING

## AB3233/3234 : loxP linker

SEQ. ID. NO. : 1

loxP site

*Bam*H I/*Bgl* II  
overhang

5' GATCCAATAACTTCGTATAGCATACATTATACGAAGTTATAAGTACTGAATTCG 3'  
3' GTTATTGAAGCATATCGTATGTAATATGCTTCAATATTTCATGACTTAAGCCTAG 5'

*Bam*H I/*Bgl* II  
overhang

SEQ. ID. NO. : 2

## AB14626/14627 : Multiple Cloning Site

SEQ. ID. NO. : 3

*Sal* I overhang

5' AATTCCCCGGGAGATCTAAGCTTGAGCTCG 3'  
3' GGGGCCCTCTAGATTCGAACTCGAGCAGCT 5'

*Eco*R I overhang

SEQ. ID. NO. : 4

## AB6920/6921 : loxP linker

SEQ. ID. NO. : 5

*Xba* I overhang

5' CTAGCAATAACTTCGTATAGCATACATTATACGAAGTTATATCGATG 3'  
3' GTTATTGAAGCATATCGTATGTAATATGCTTCAATATAGCTACGATC 5'

*Xba* I overhang

SEQ. ID. NO. : 6

## AB14680/14681 : loxP linker

SEQ. ID. NO. : 7

*Blp* I overhang

5' TGACAATAACTTCGTATAGCATACATTATACGAAGTTATATCGATG 3'  
3' GTTATTGAAGCATATCGTATGTAATATGCTTCAATATAGCTACACT 5'

*Blp* I overhang

SEQ. ID. NO. : 8

Fig. 3

# CONSTRUCTION OF A CIRCULAR GENOMIC PLASMID FOR Ad VECTOR RESCUE USING THE Cre/ loxP SYSTEM

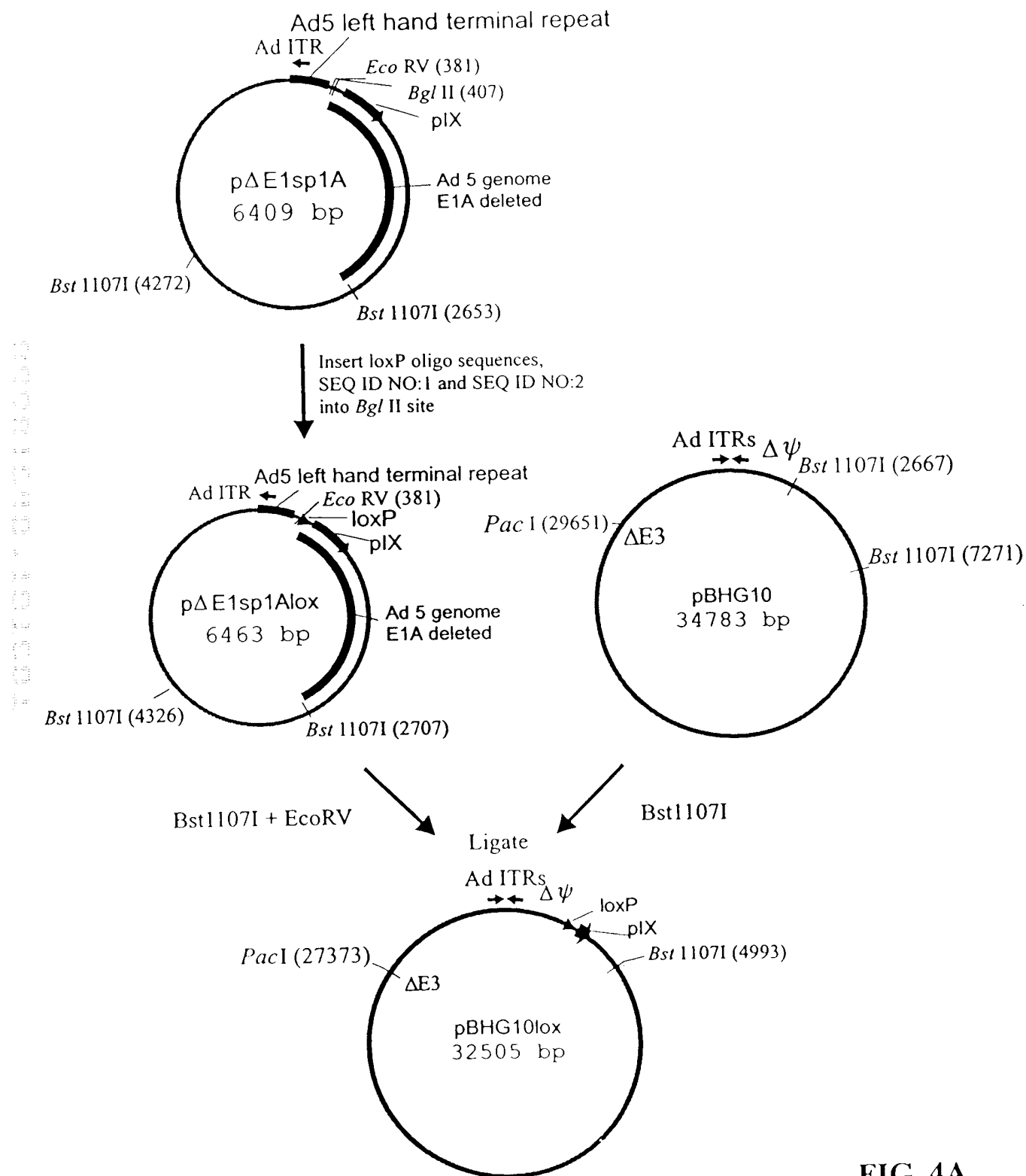
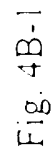


FIG. 4A

[illegible]

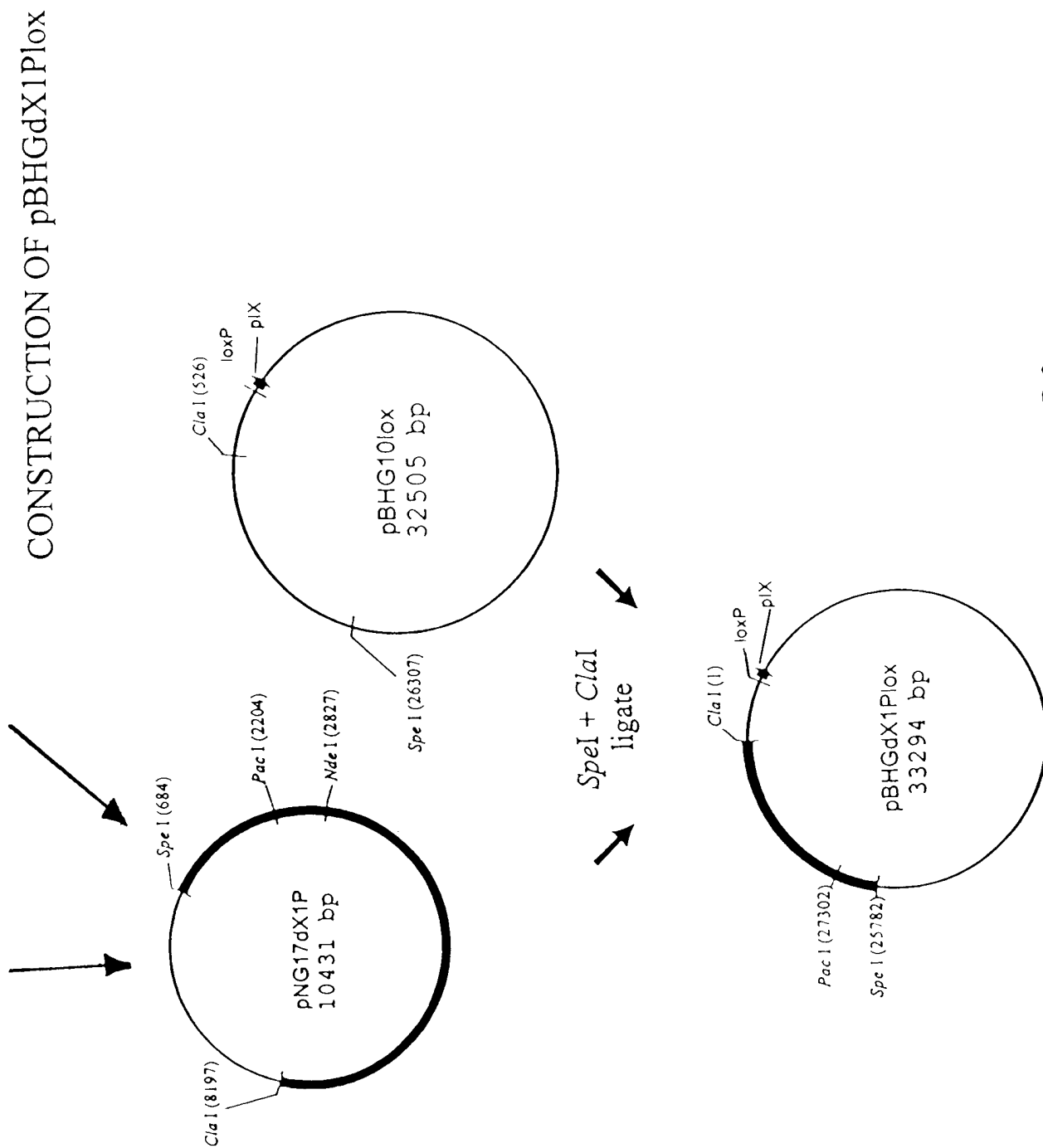


Fig. 4B-2

# CONSTRUCTION OF pBHGE3lox

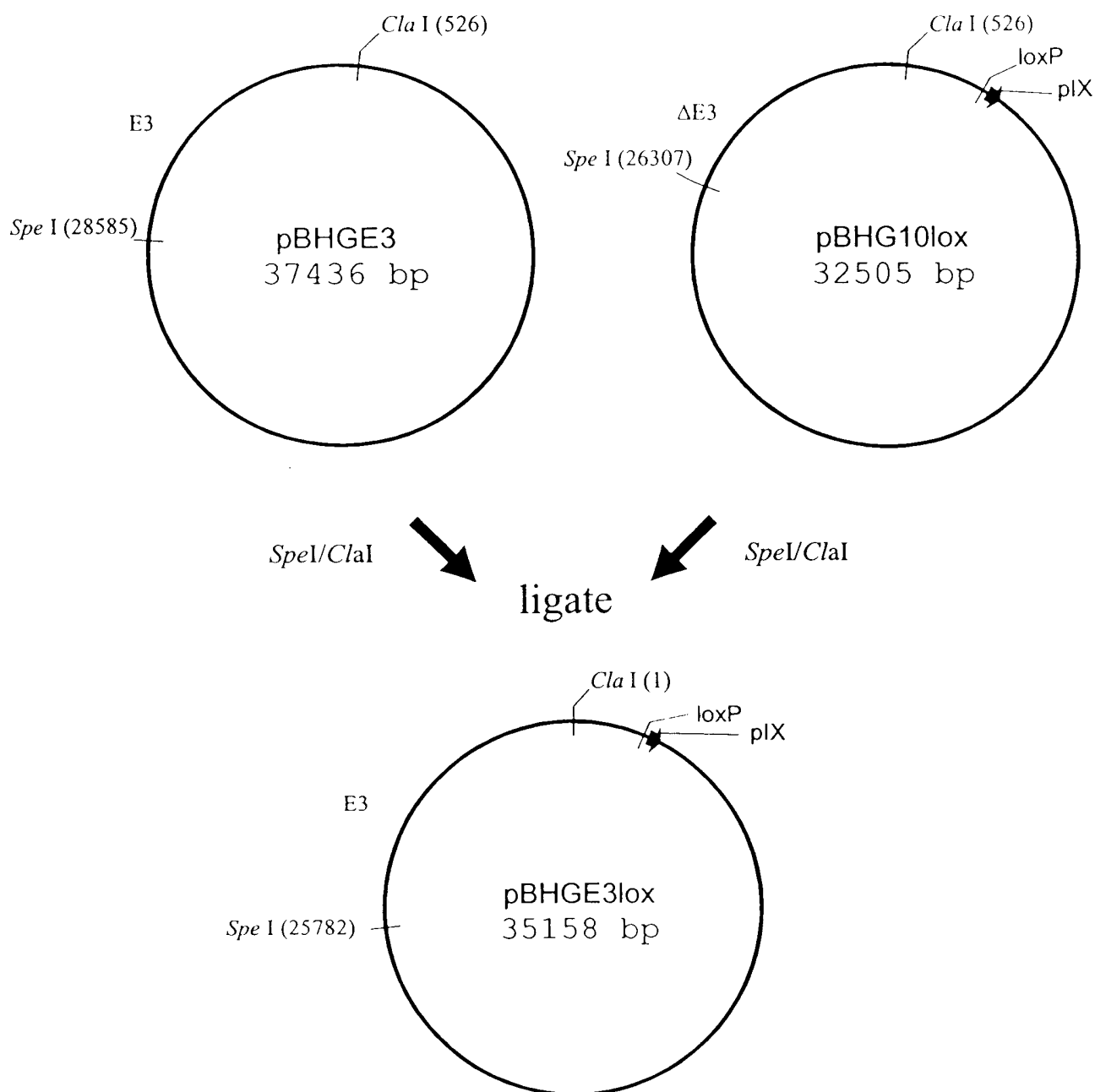


Fig. 4C



# CONSTRUCTION OF Ad GENOMIC PLASMIDS ENCODING CRE

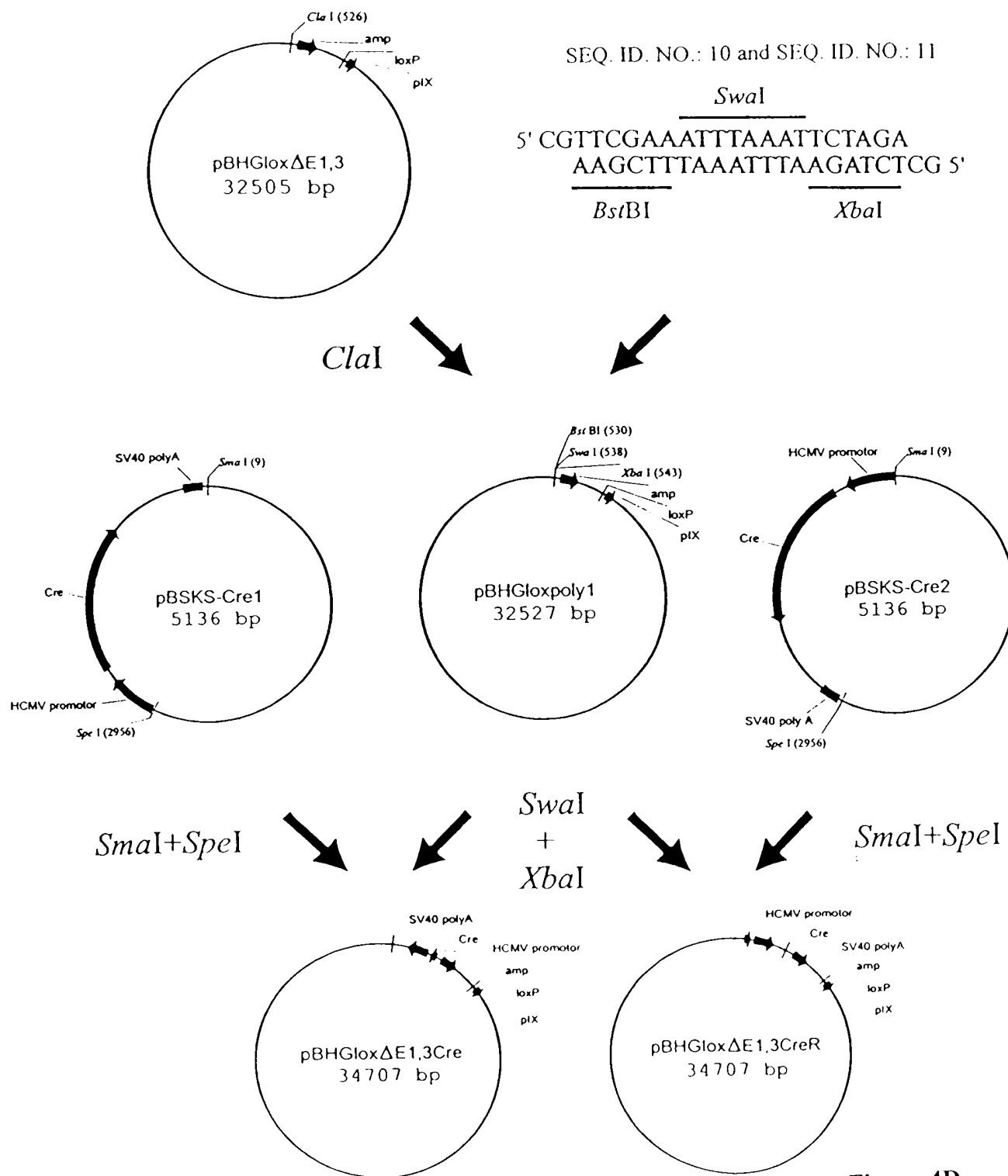


Figure 4D

# CONSTRUCTION OF pΔE1SP1A & pΔE1SP1B loxP PLASMIDS FOR RESCUE OF FOREIGN DNA

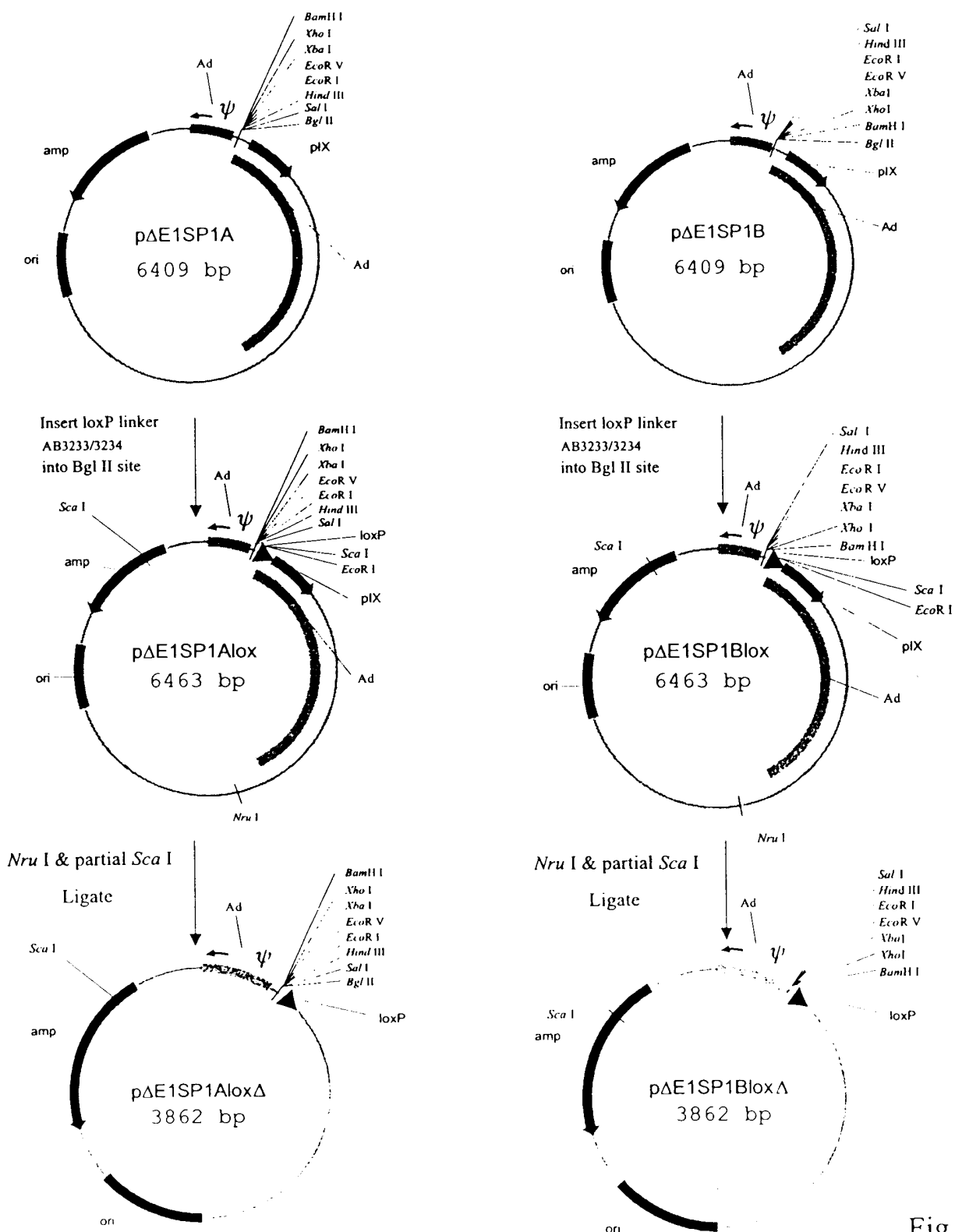
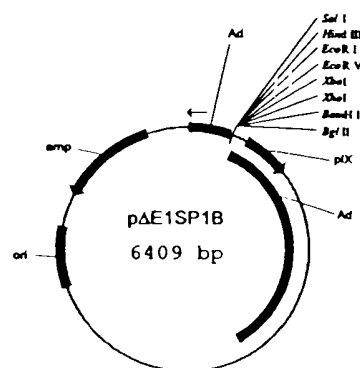
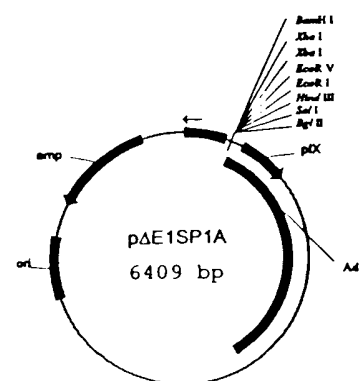
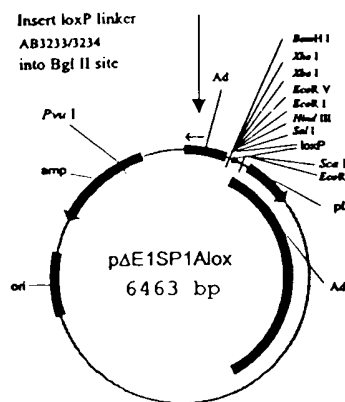


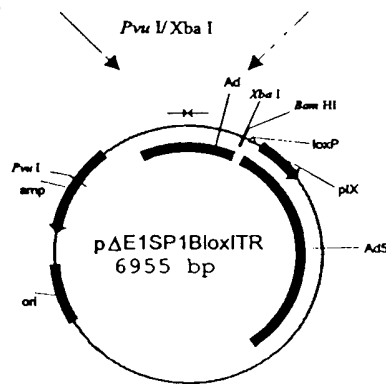
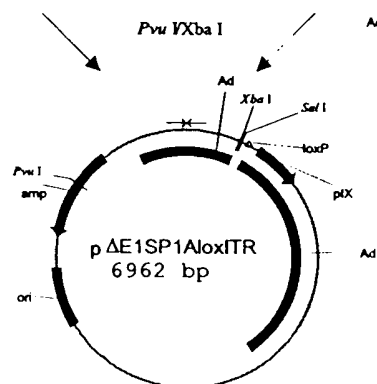
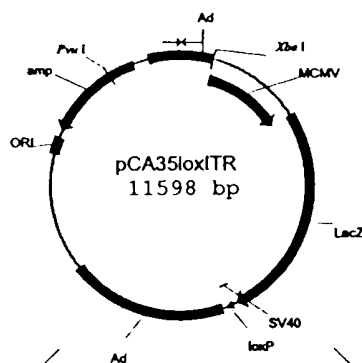
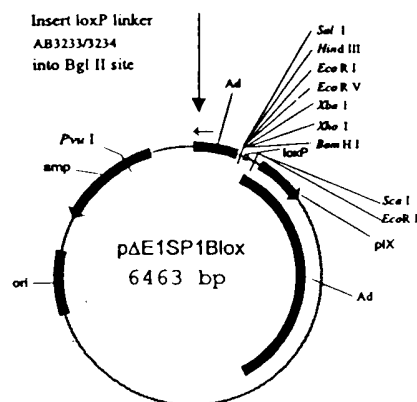
Fig. 5A



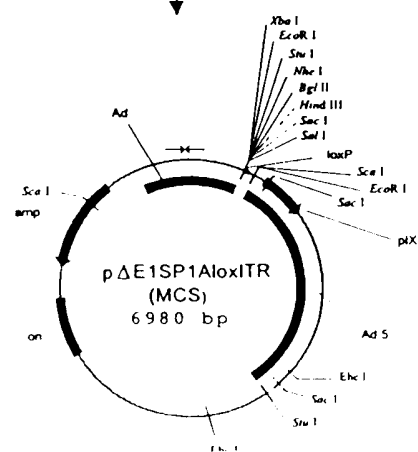
Insert loxP linker  
AB3233/3234  
into Bgl II site



Insert loxP linker  
AB3233/3234  
into Bgl II site



Xba I/Sal I  
AB16853/16854



Xba I/BamHI  
AB 16855/16856

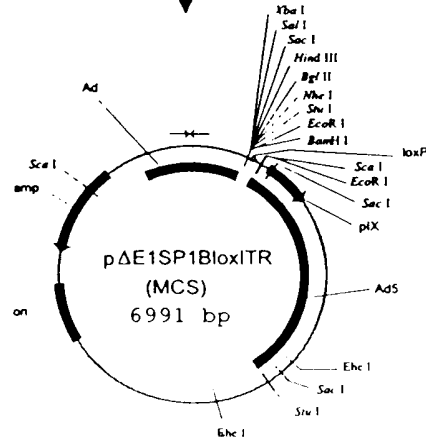


Figure 5B

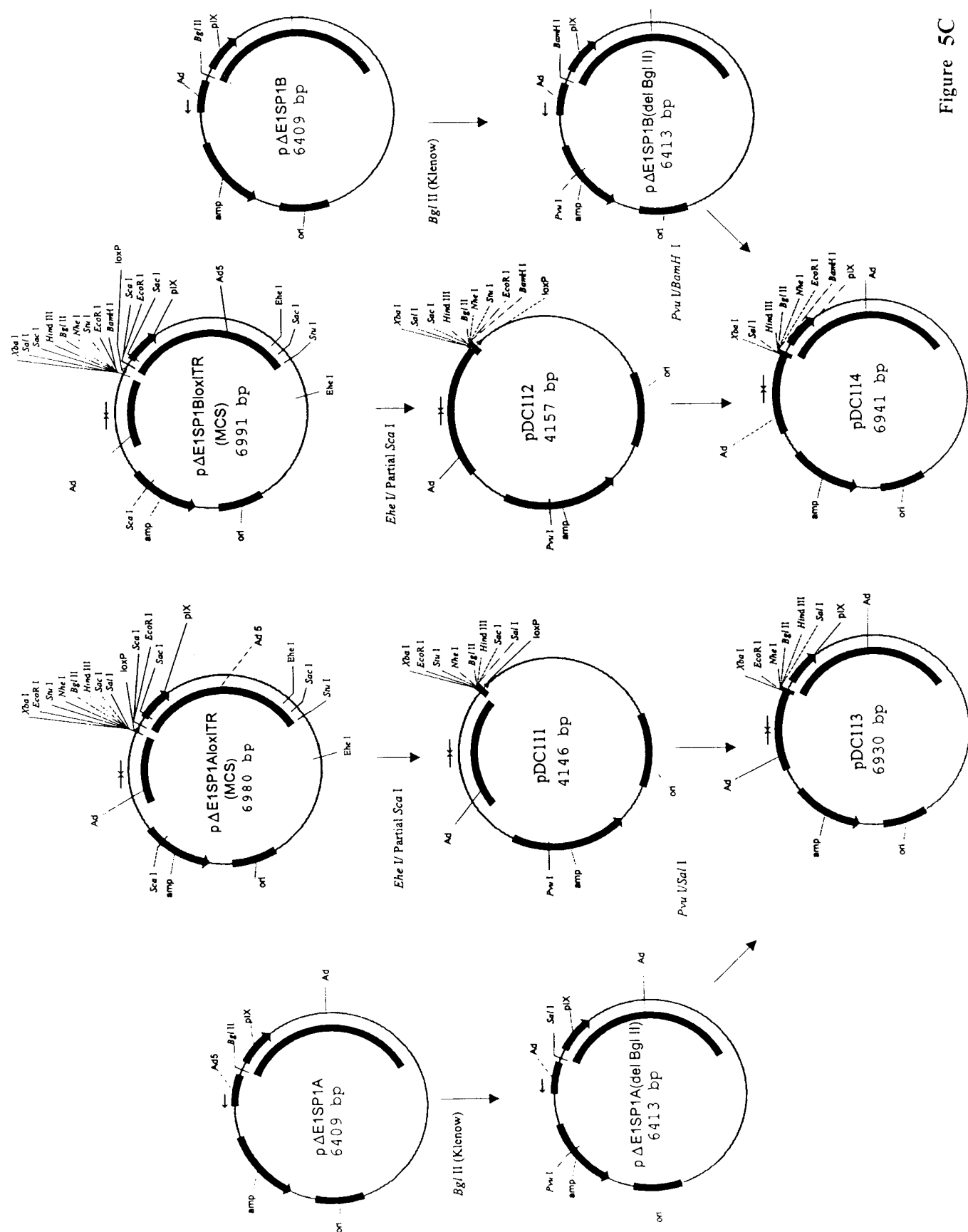
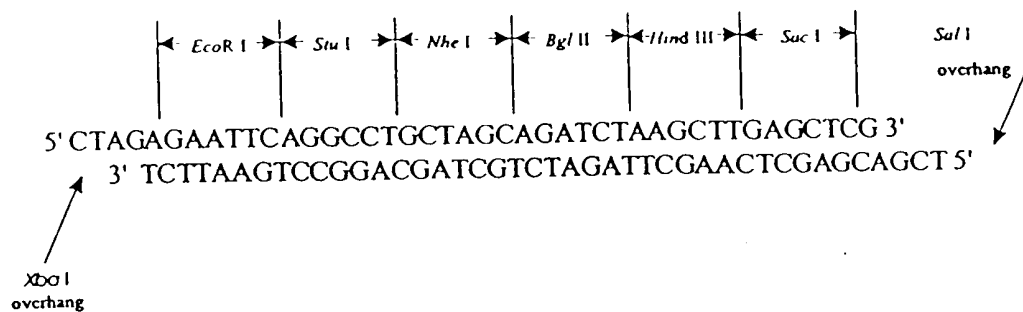


Figure 5C

SEQ. ID. NO.: 12 (AB16853) and SEQ. ID. NO.: 13 (AB16854)



SEQ. ID. NO.: 14 (AB16855) and SEQ. ID. NO.: 15 (AB16856)

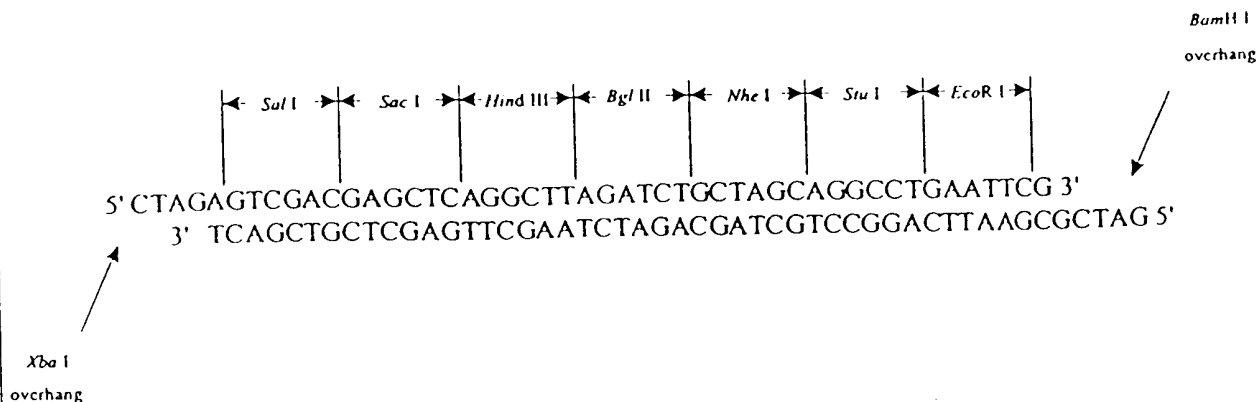


Figure 5D

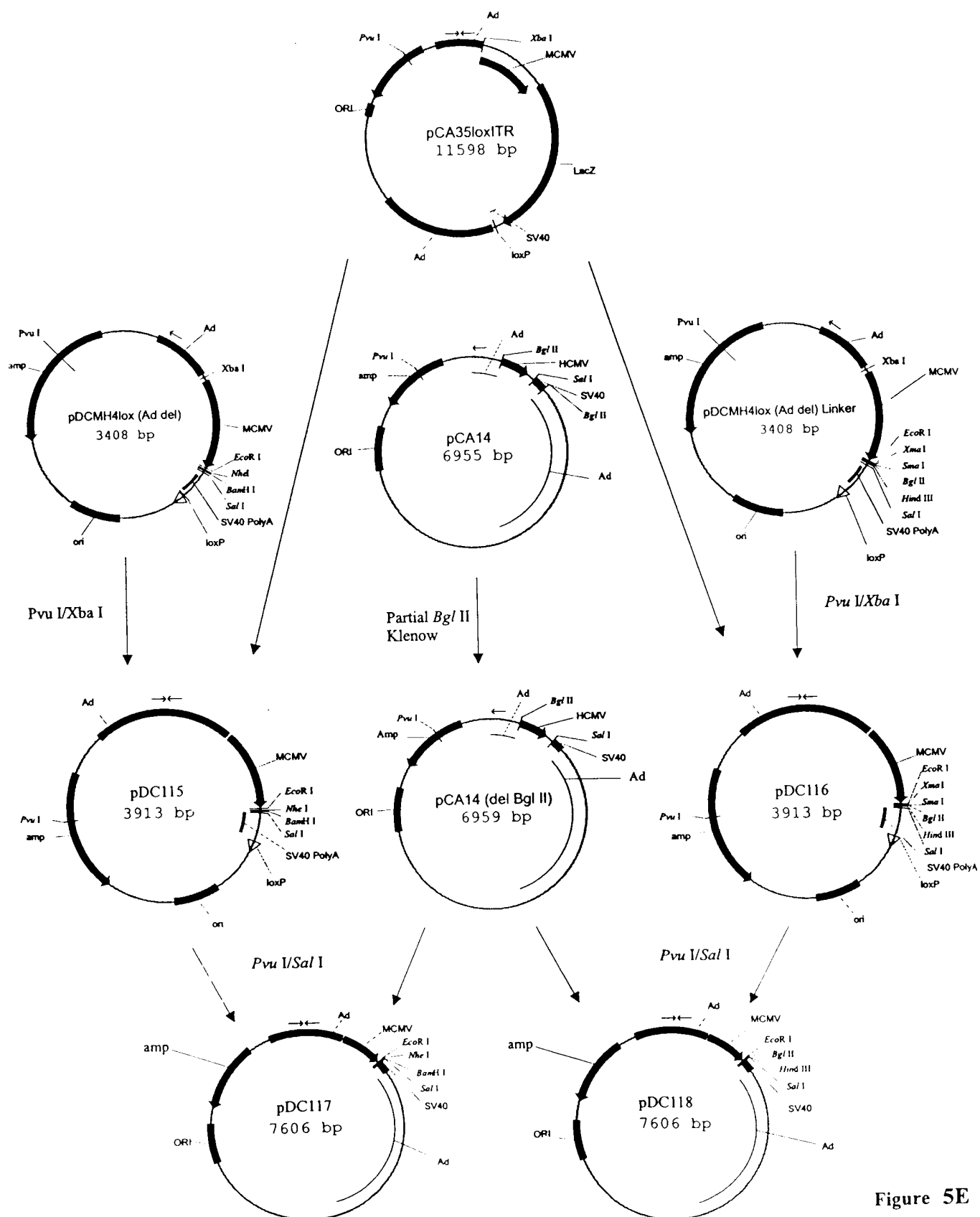


Figure 5E

# CONSTRUCTION OF pMH4LOX, pMH4LOX $\Delta$ and pMH4LOX $\Delta$ LINK SHUTTLE PLASMIDS FOR RESCUE OF EXPRESSION CASSETTES

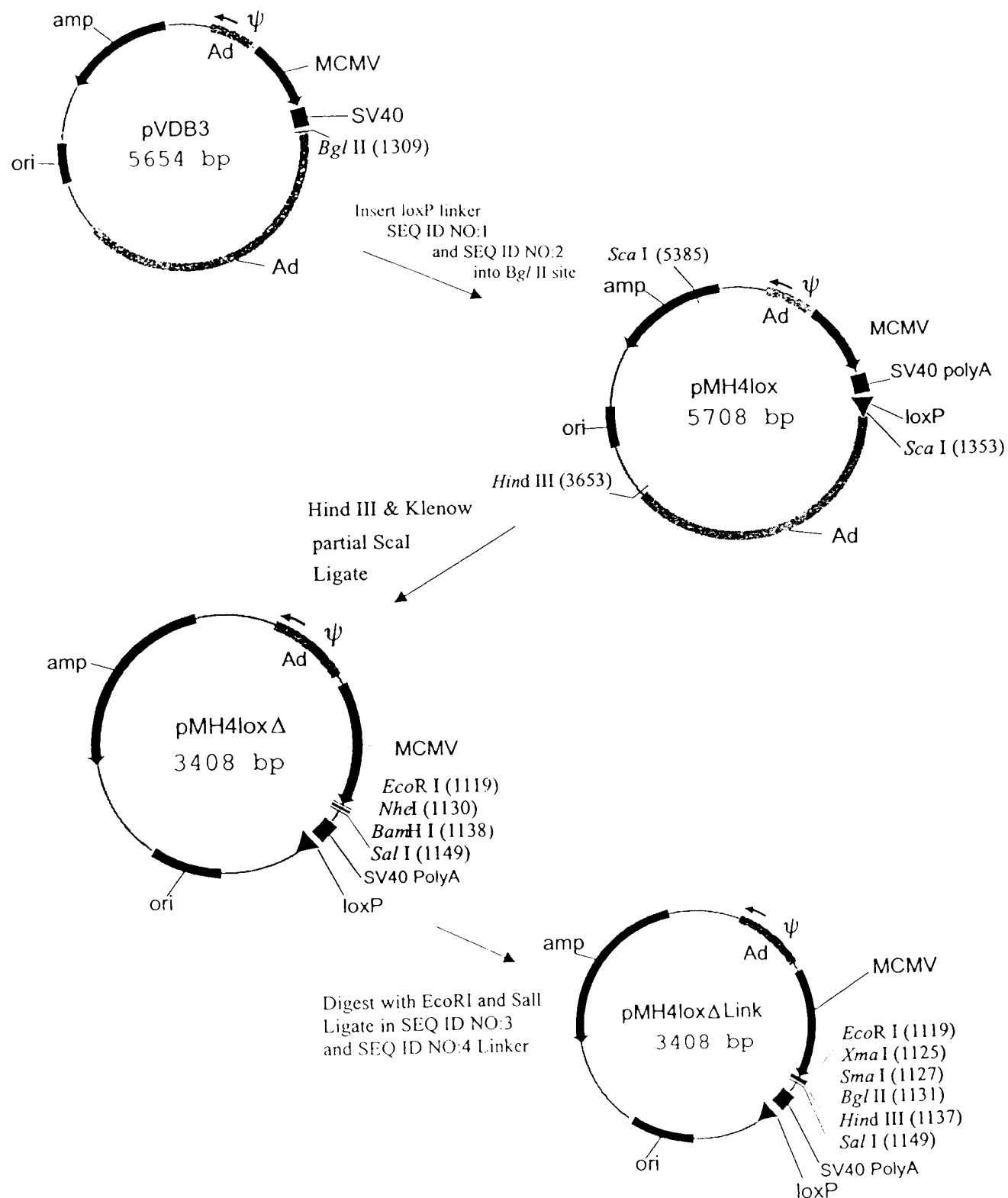


Fig. 6A

# CONSTRUCTION OF A SHUTTLE PLASMID CONTAINING A pUC DERIVED ORIGIN

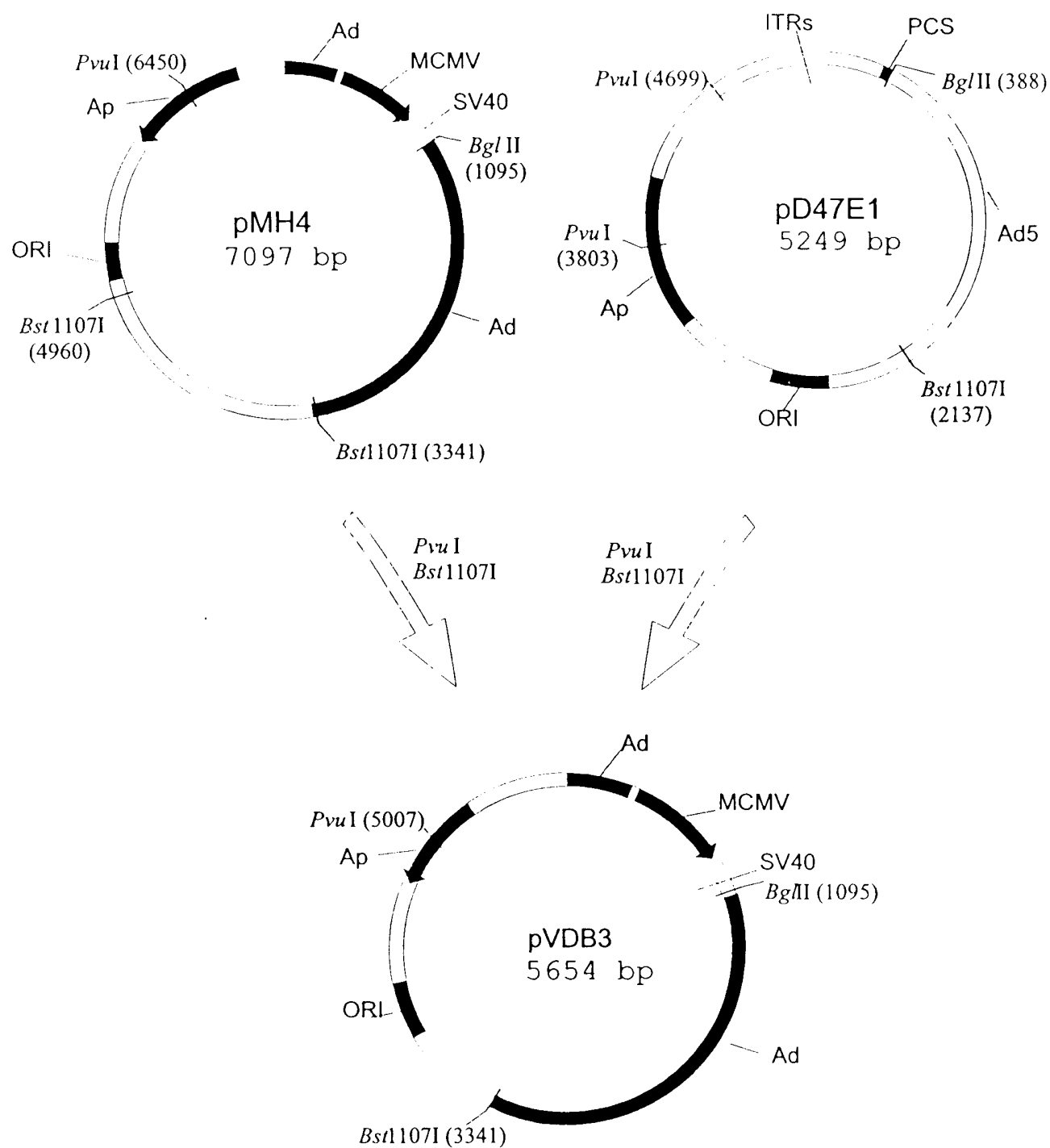
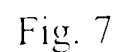


Fig. 6B



[illegible]

# CONSTRUCTION OF pCA36LOX and pCA36LOX $\Delta$ SHUTTLE PLASMIDS FOR RESCUE OF LACZ

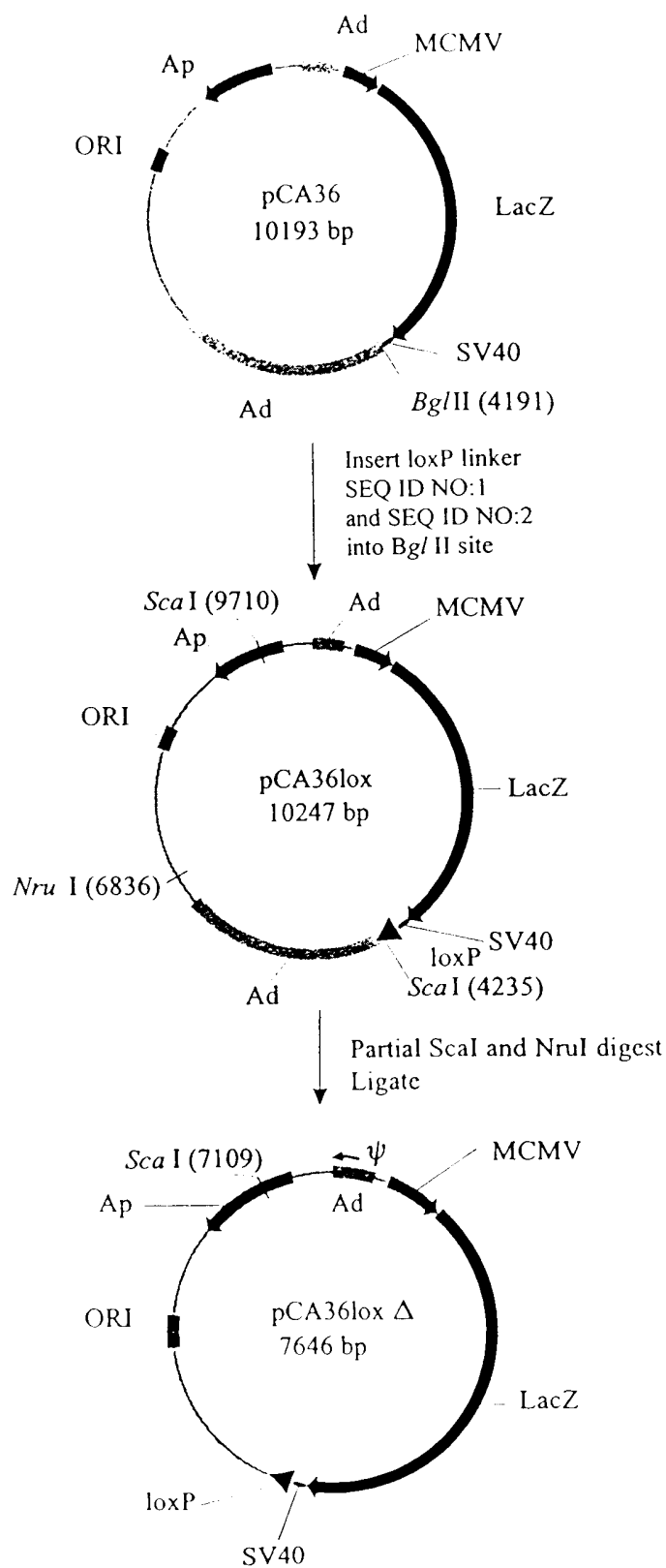


Fig. 8A

Cotransfection of 293Cre cells with AdLC8c DNA-TP and a shuttle plasmid containing a loxP site for generation of Ad expression vectors

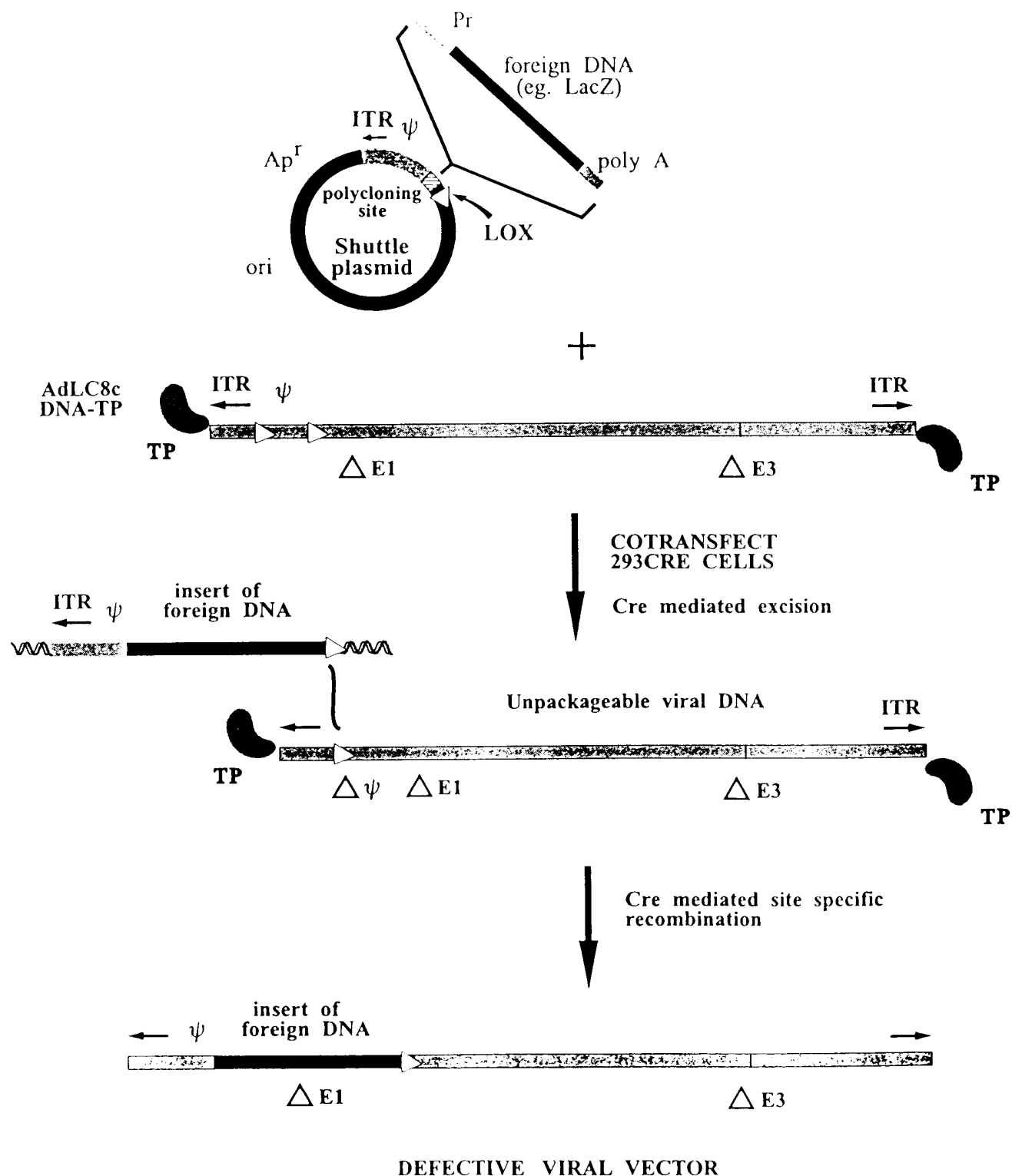


Fig. 8B

# Cotransfection of 293Cre cells with restricted AdLC8c DNA-TP and loxP shuttle plasmid for generation of Ad expression vectors

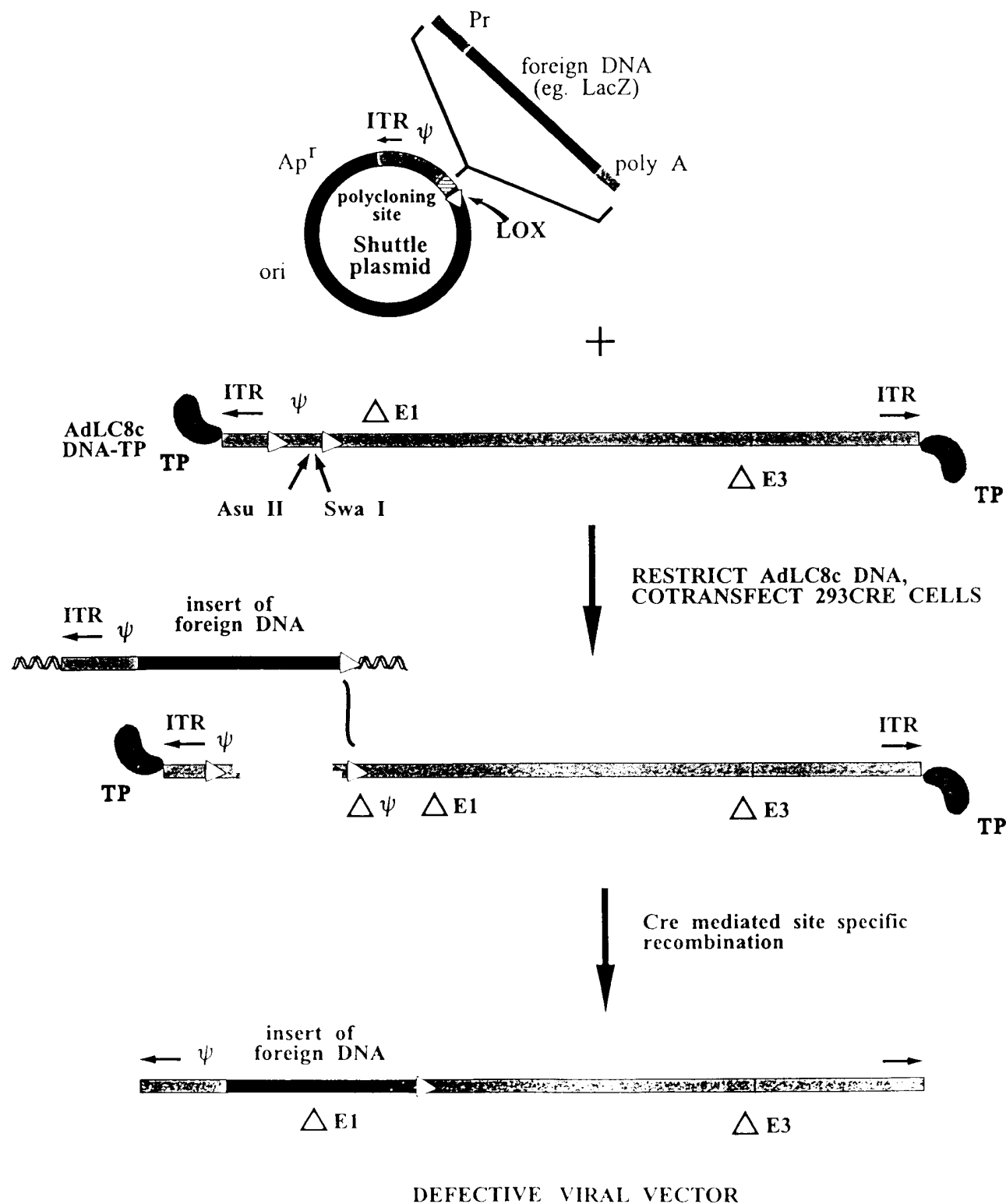


Fig. 8C

# CONSTRUCTION OF SHUTTLE PLASMIDS EXPRESSING Cre

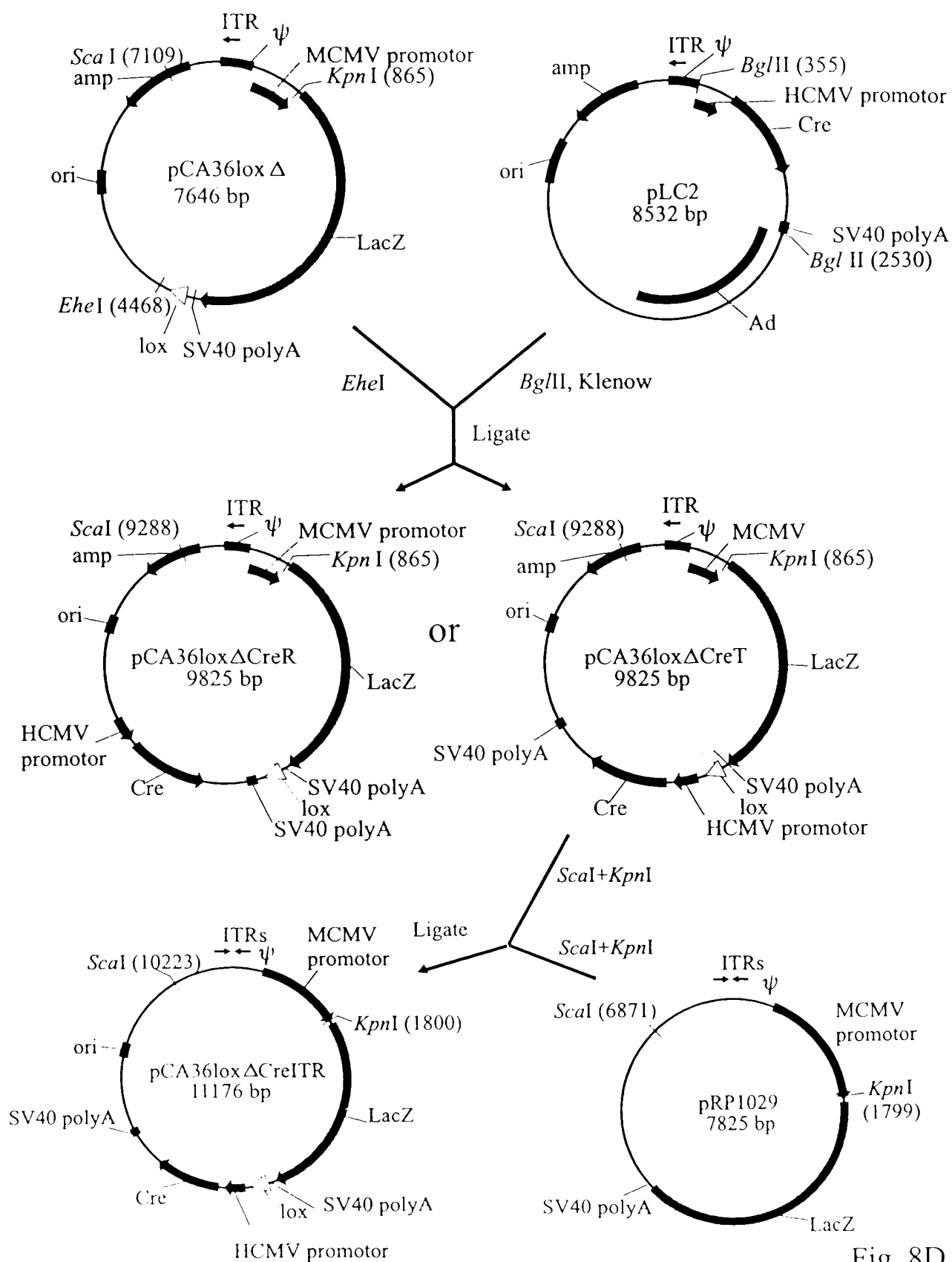


Fig. 8D

Cotransfection of 293 cells with pBHG10lox and a "Lox" shuttle plasmid expressing Cre for generation of Ad expression vectors

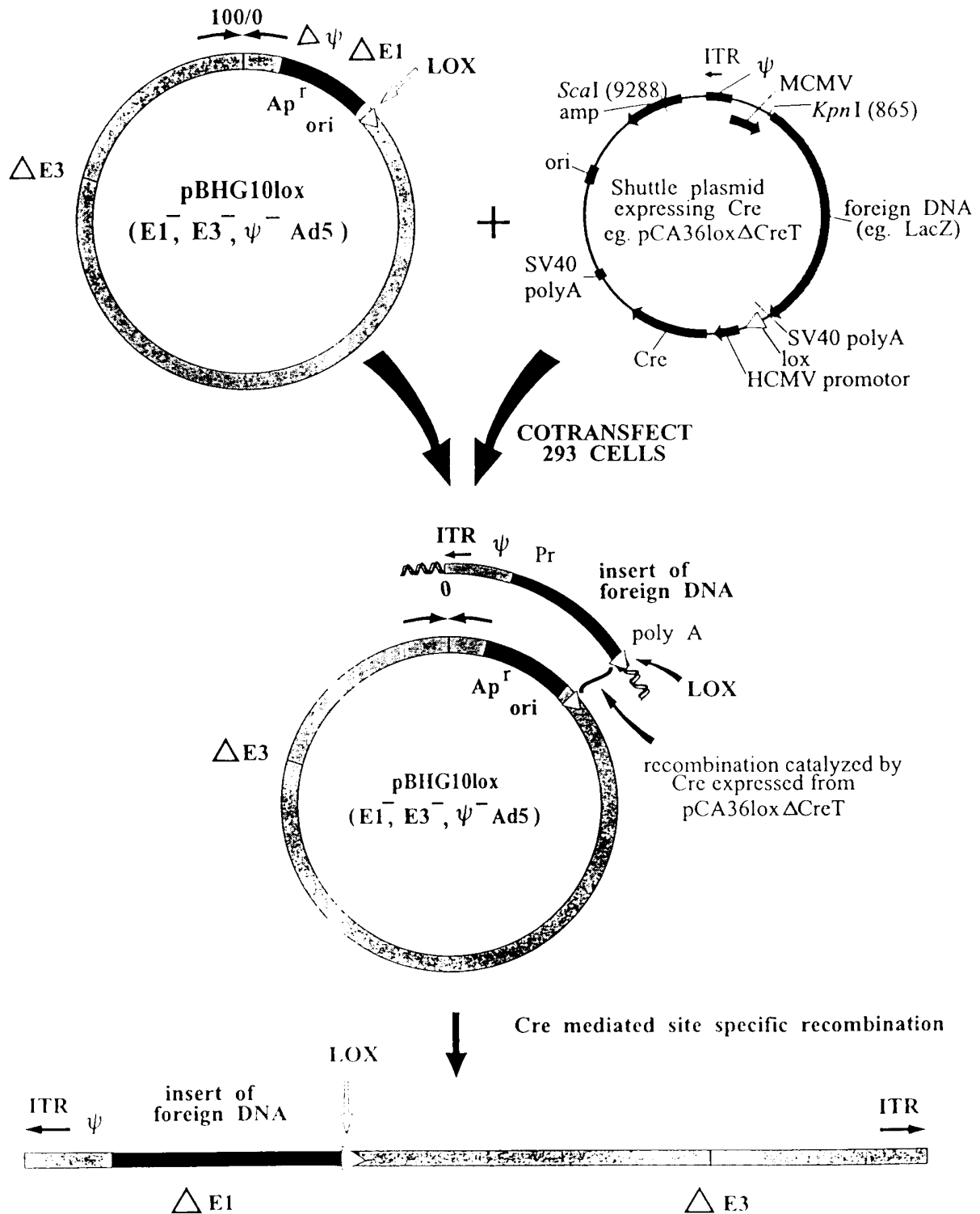


Fig. 8E

# CONSTRUCTION OF Ad GENOMIC PLASMID ENCODING CRE

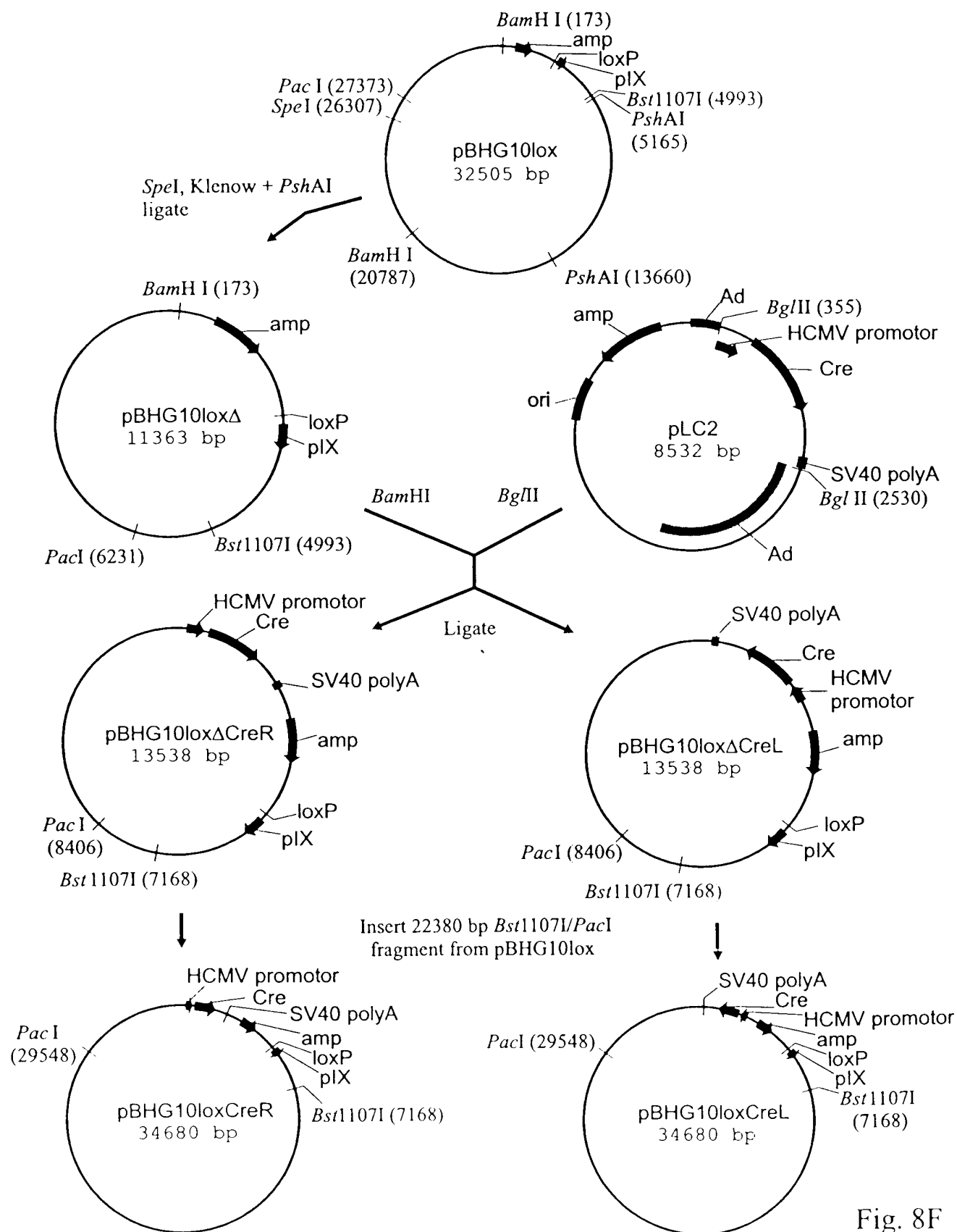
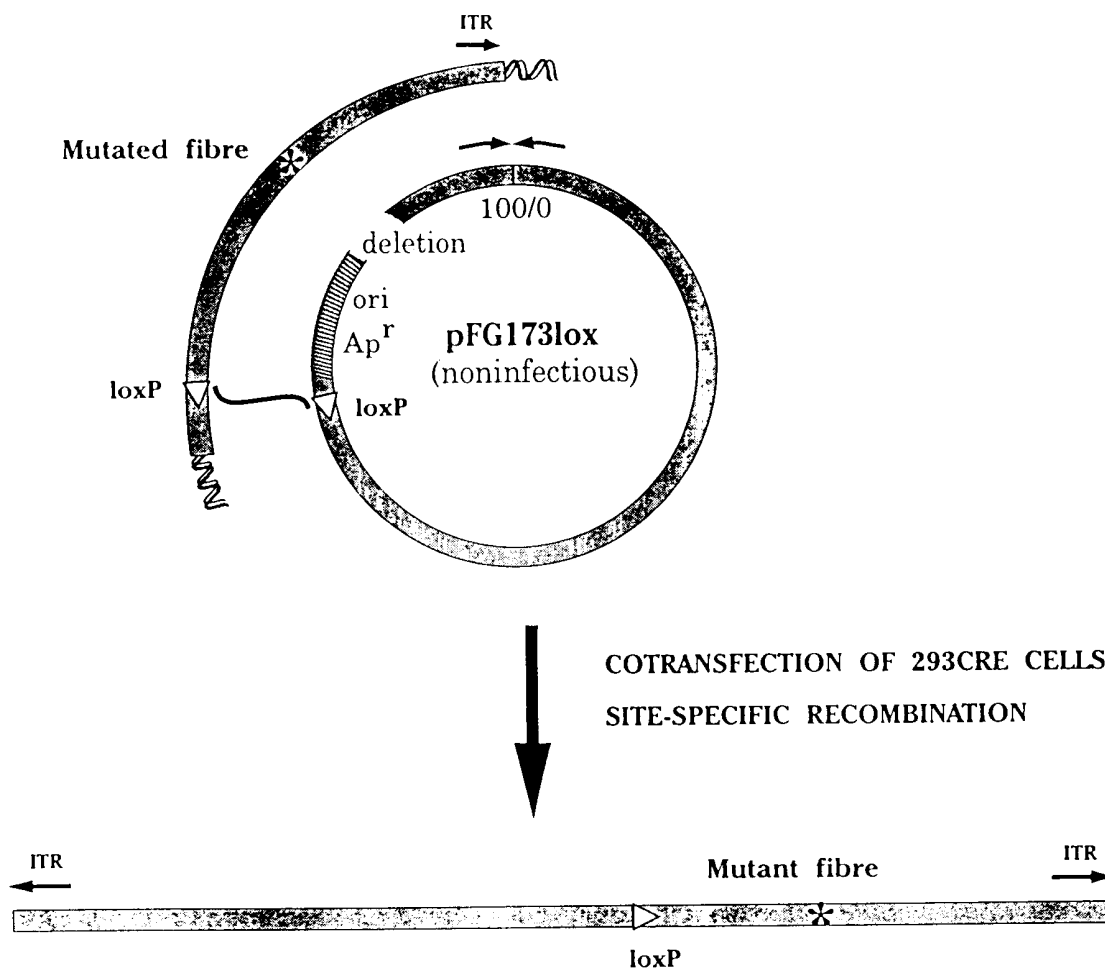


Fig. 8F

# RESCUE OF FIBRE MUTATIONS USING CRE/LOX RECOMBINATION



NONDEFECTIVE (E1<sup>+</sup>) VIRUS WITH MUTATED FIBRE GENE

Fig. 9A



# CONSTRUCTION OF pAB14lox $\Delta$

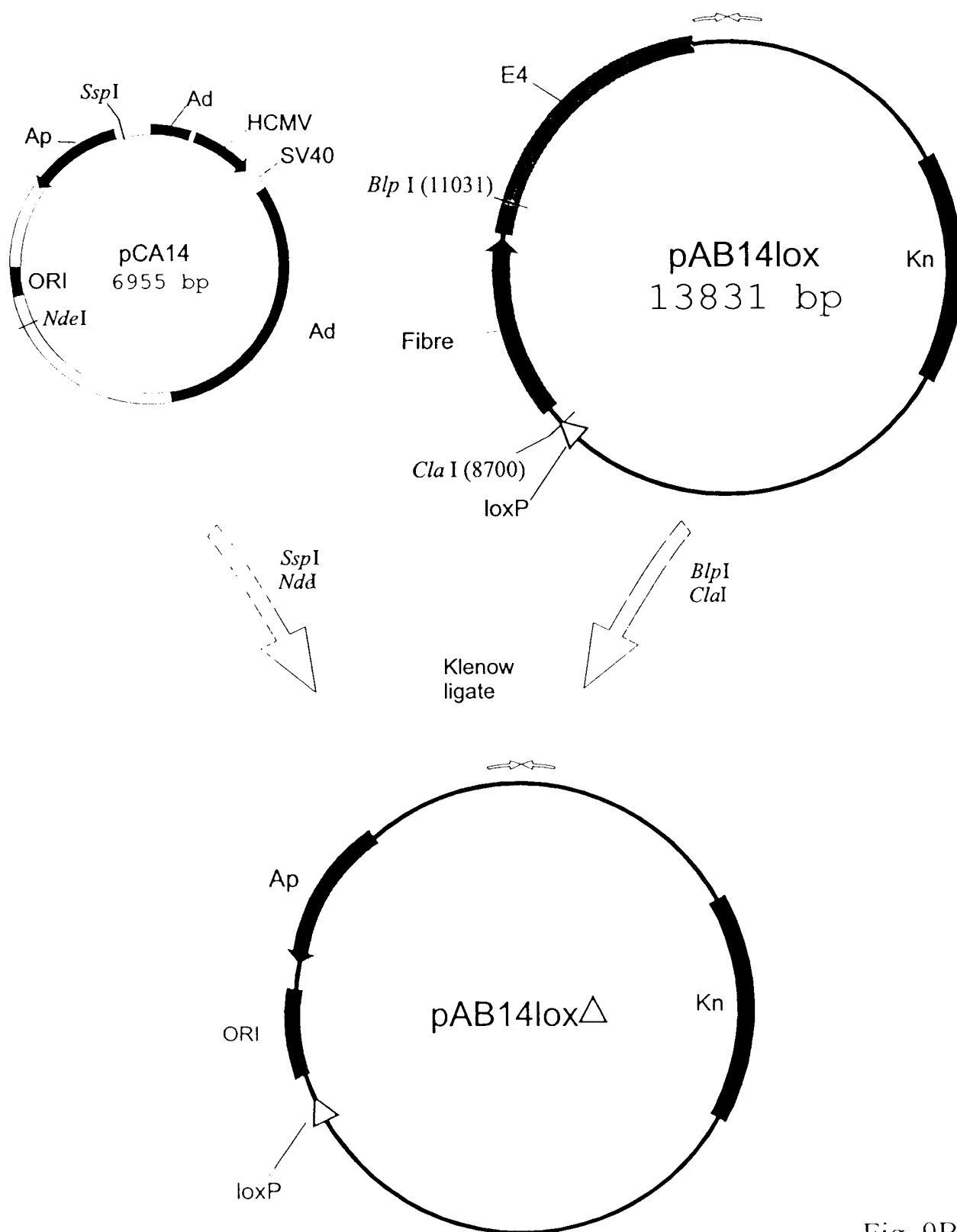
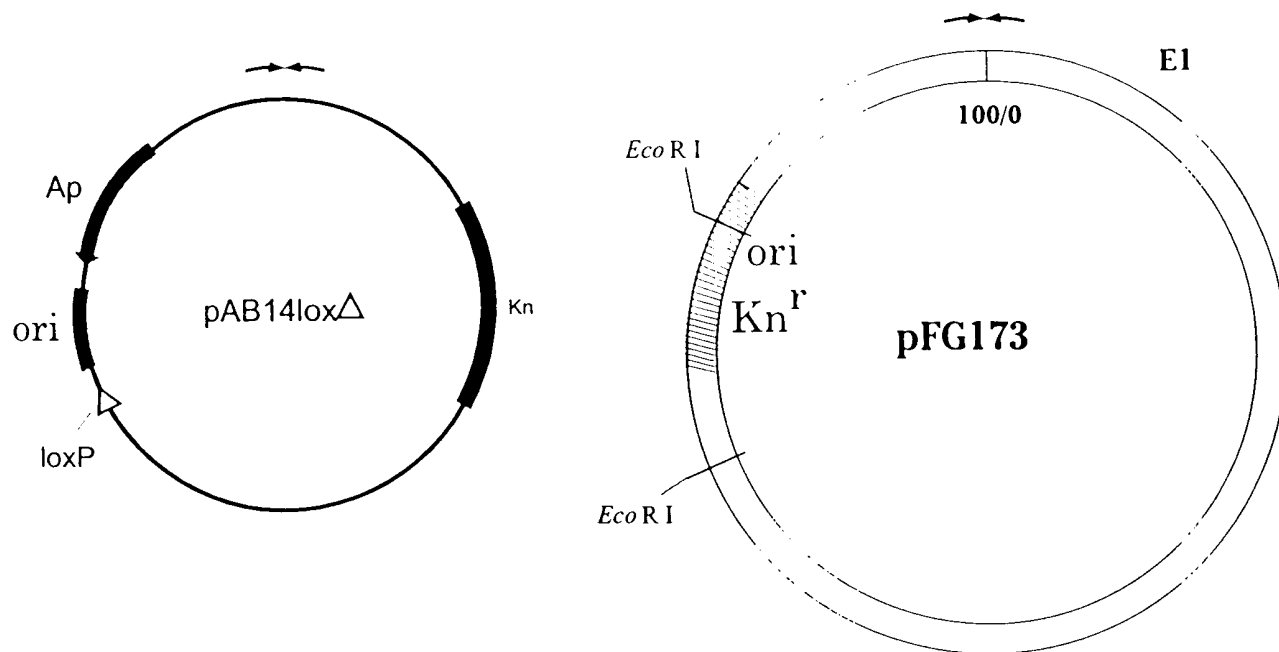


Fig. 9B

# CONSTRUCTION OF pFG173lox



Restriction, transformation of *E. coli*,  
homologous recombination

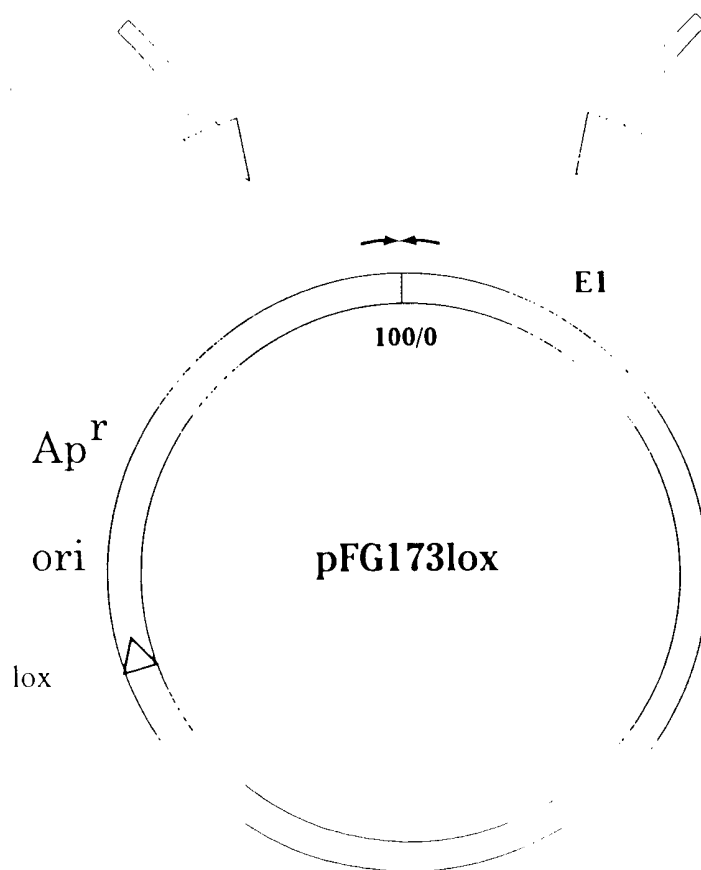


Fig. 9C

# CONSTRUCTION OF pFG23dX1lox AND pFG23dX1loxc FOR RESCUE OF MUTANT FIBRE INTO AD VIRUS

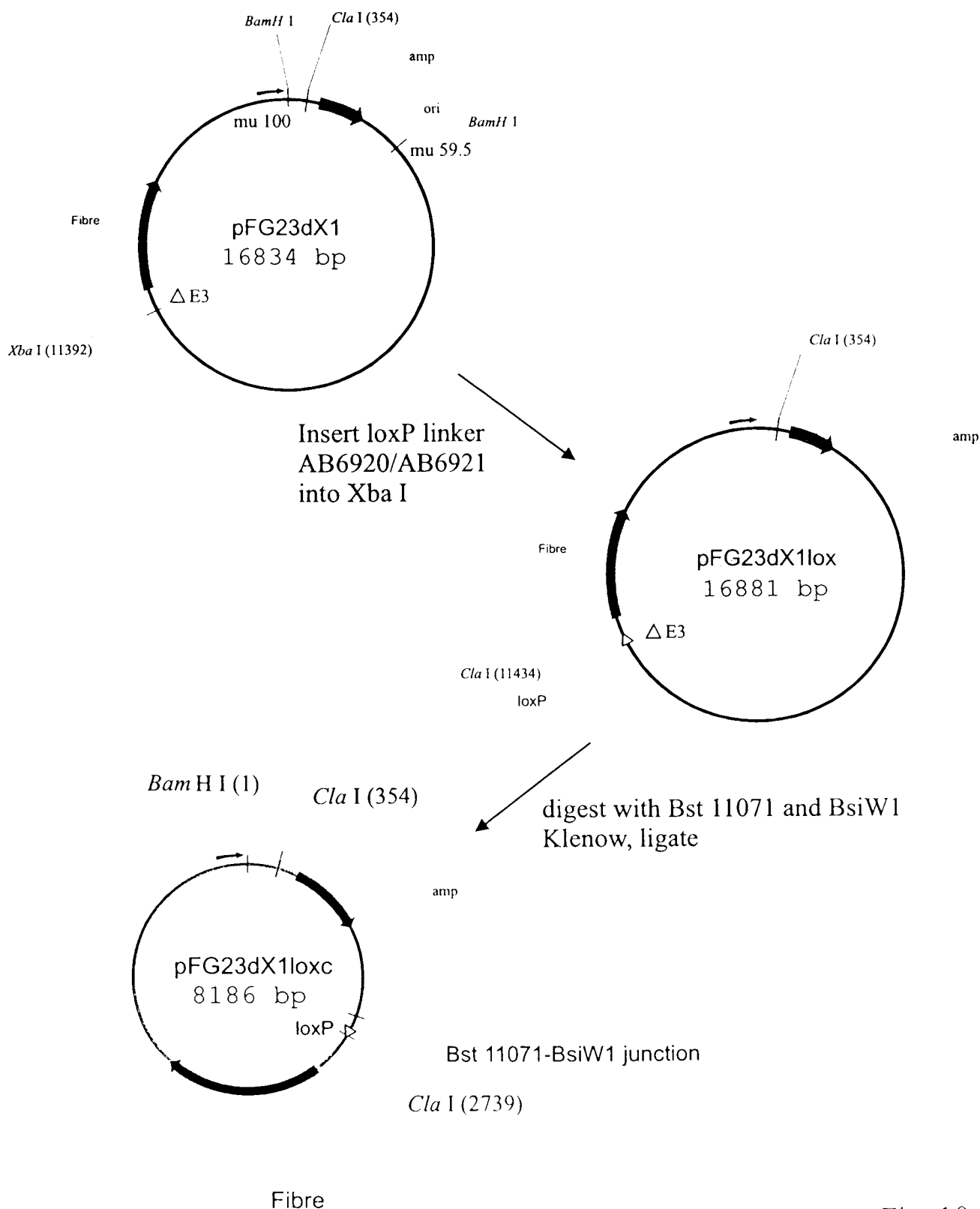


Fig. 10

# A PLASMID FOR RESCUE OF A FOREIGN DNA INTO AD VIRUS

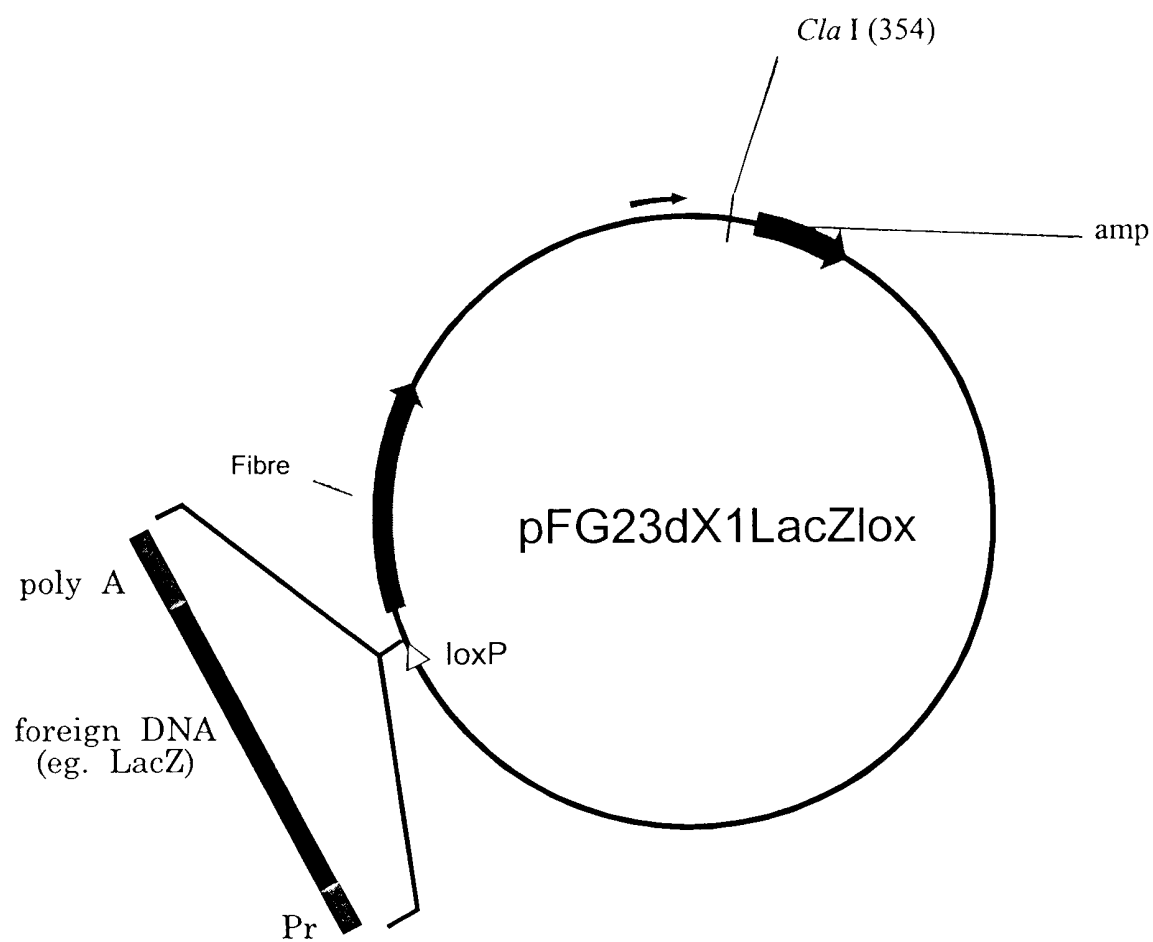
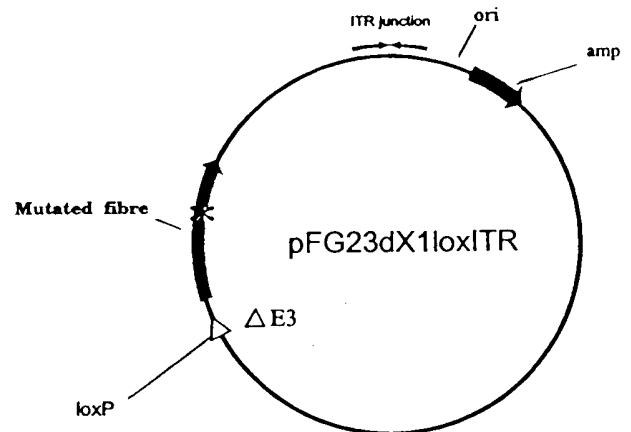
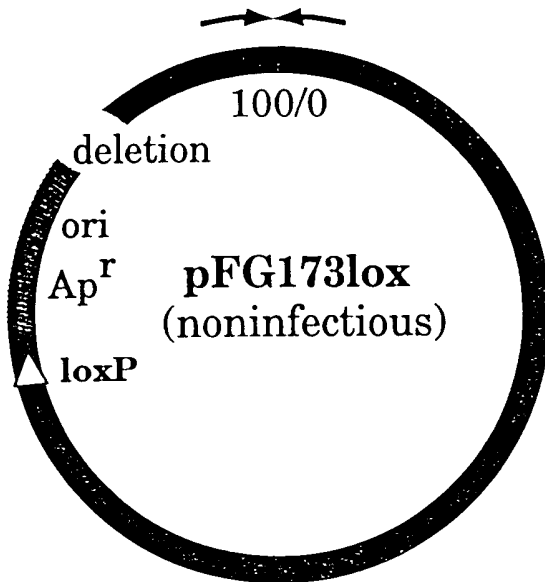


Fig. 11A

# RESCUE OF FIBRE MUTATIONS USING CRE/LOX RECOMBINATION



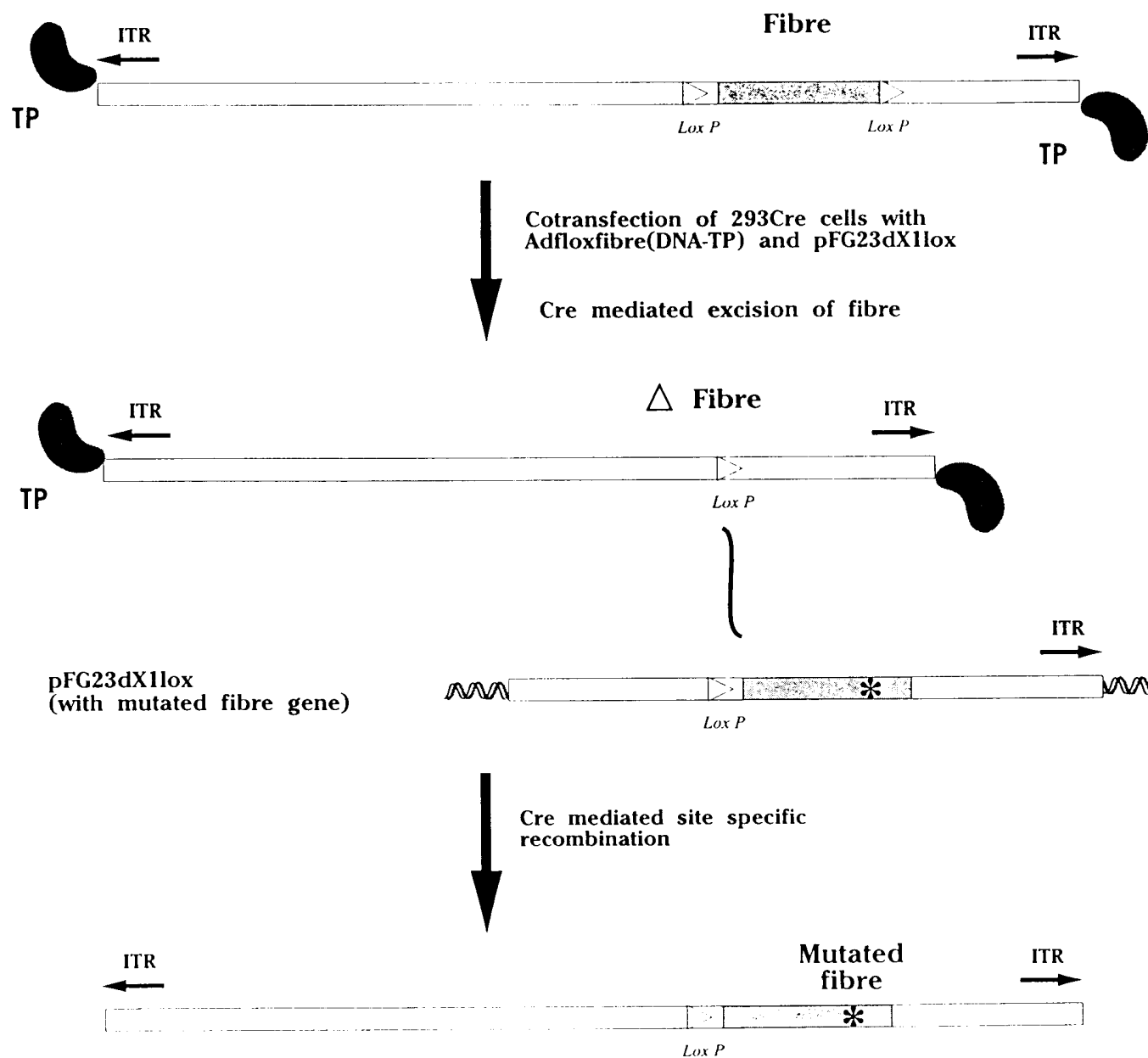
COTRANSFECTION OF 293CRE CELLS  
SITE-SPECIFIC RECOMBINATION



NONDEFECTIVE (E1<sup>+</sup>) VIRUS WITH MUTATED FIBRE GENE

FIGURE 11B

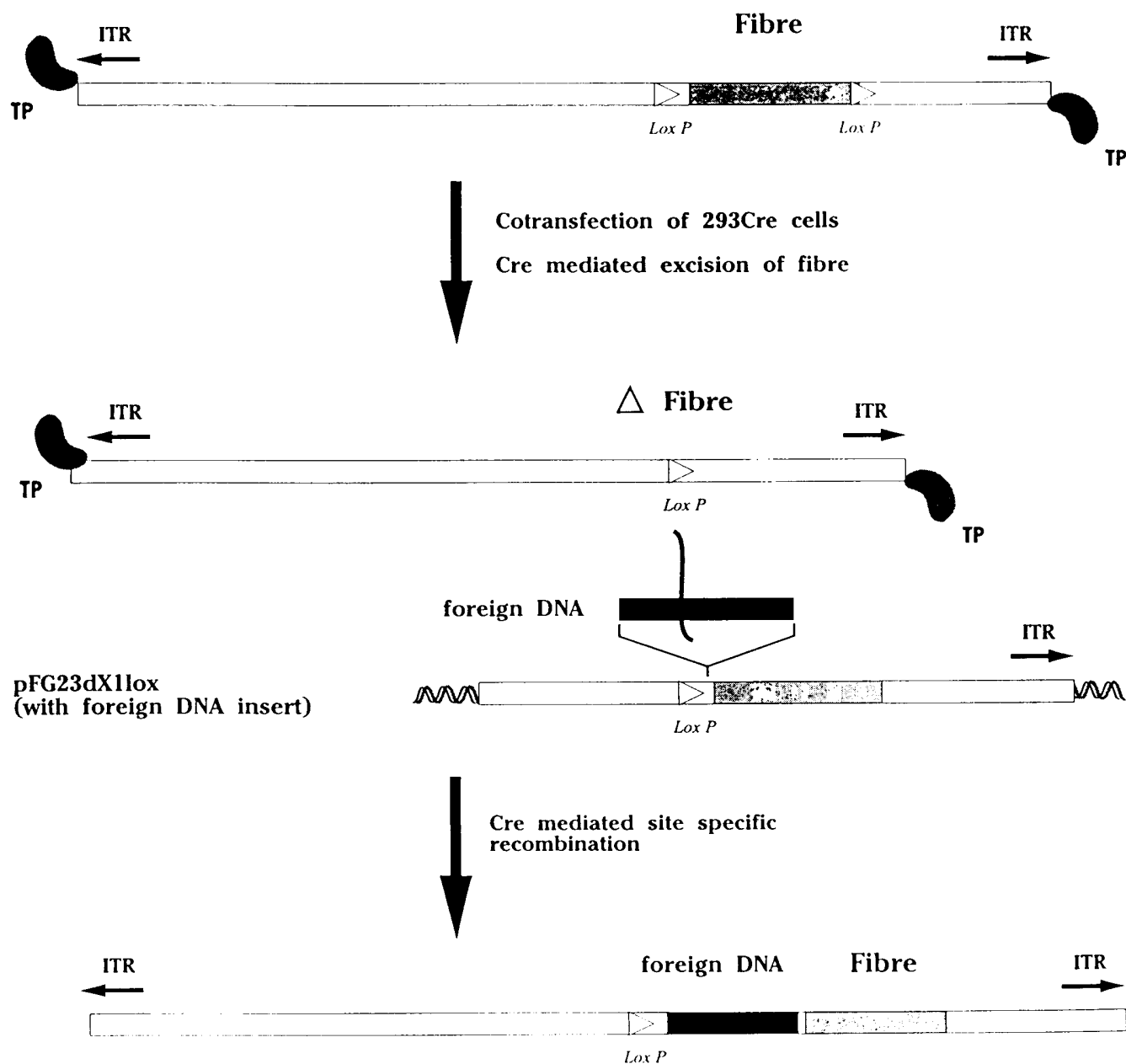
# Isolation of a virus containing a mutant fibre gene by Cre-lox recombination using DNA-TP and cotransfection



RECOMBINANT VIRUS CONTAINING A MUTATED FIBRE GENE

Fig. 12

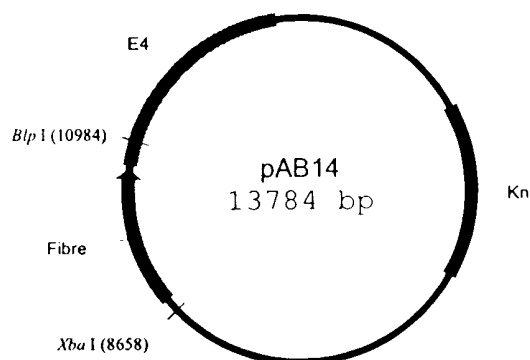
# Isolation of a virus containing a foreign DNA insert upstream of the fibre gene by Cre-lox recombination



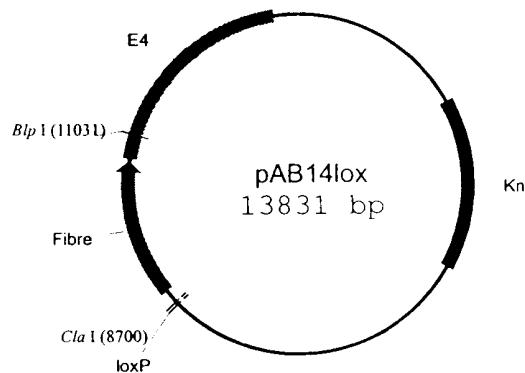
RECOMBINANT VIRUS CONTAINING AN INSERT OF FOREIGN DNA  
UPSTREAM OF THE FIBRE GENE

Fig. 13

# CONSTRUCTION OF pAB14FL0X FOR ISOLATION OF AN AD VIRUS WITH A FLOXED FIBRE GENE



Insert loxP linker  
AB6920/AB6921  
into Xba I site



Insert loxP linker  
AB14680/AB14681  
into Bsp 1 site

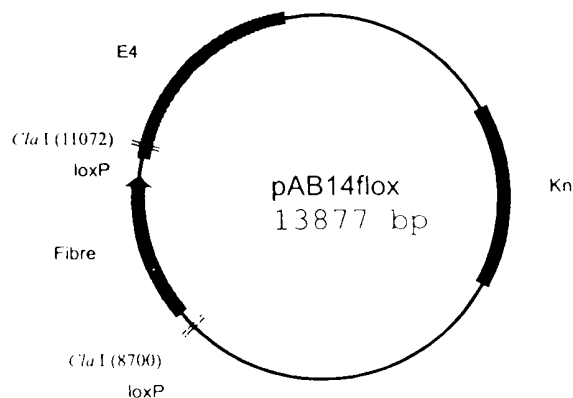
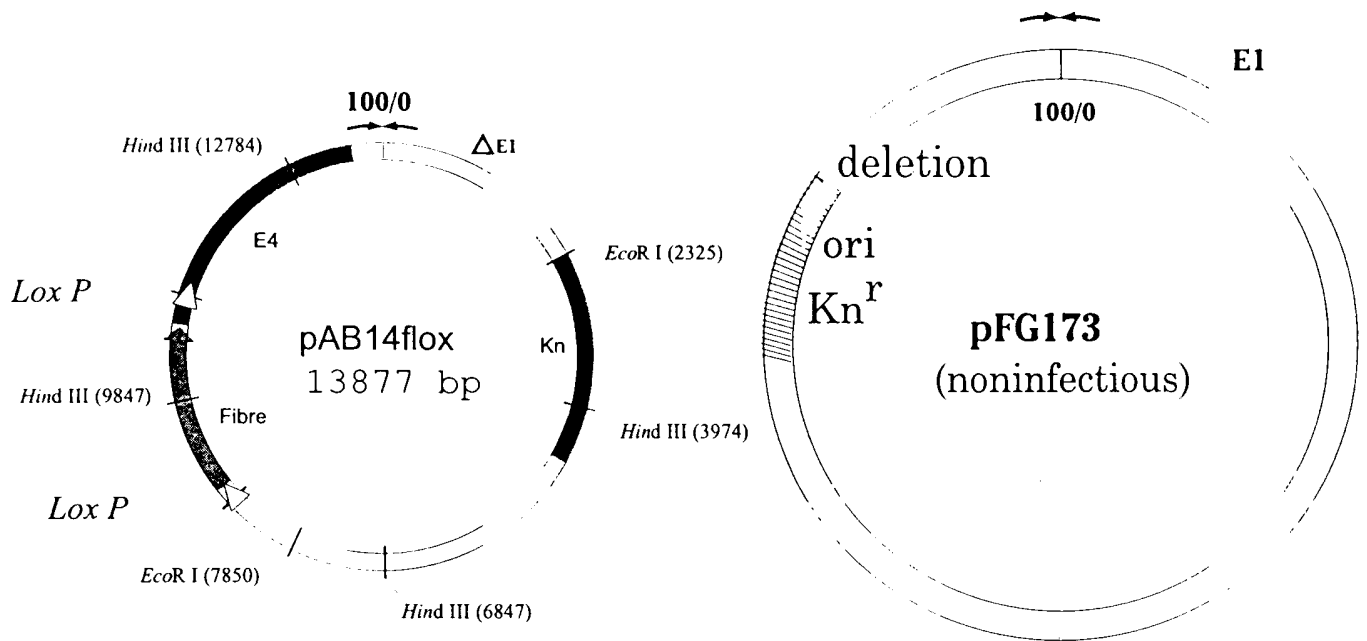


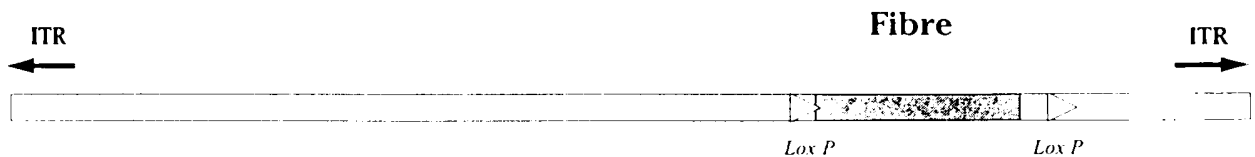
Fig. 14



# Isolation of a virus containing a fibre gene with flanking lox P sites.



COTRANSFECTION OF 293 CELLS  
HOMOLOGOUS RECOMBINATION



NONDEFECTIVE (E1<sup>+</sup>) VIRUS (ADFLOXFIBRE) CONTAINING A FLOXED FIBRE GENE

Fig.15